

APPARATUS AND TECHNIQUE FOR PHOTOGRAPHING GRAIN KERNELS AND SIMILAR OBJECTS<sup>1</sup>V. G. MARTIN<sup>2</sup> AND J. ANSEL ANDERSON<sup>3</sup>*Grain Research Laboratory, Board of Grain Commissioners for Canada, Winnipeg, Manitoba*

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Photographs of grain kernels and other small objects, which are to be reproduced in scientific journals, require special equipment and technique. They are too small for ordinary cameras, and a microscope with camera attachment is unsuitable because of limitations in the depth and area of the field. Commercial equipment for low-power photography is available, but is relatively expensive. This paper describes a simple camera, built in this laboratory, for producing photographs of specimens of actual size, or magnifications up to about four diameters. Information on accessory equipment, films, developer, colour photography, and technique, is also included.

## EQUIPMENT AND TECHNIQUE

*Camera*

A drawing and a photograph of the camera are shown in Figures 1 and 2. The body of the camera consists of a bronze cone cast from wood patterns made in the laboratory workshop. Lens and shutter were purchased and are mounted in a tube which threads into the lower end of the cone. The square threads have a lead of  $\frac{1}{8}$  inch per revolution and thus provide for rapid and accurate focusing. A bakelite adapter, screwed to the top of the cone and lined with plush, takes either a ground glass focusing panel or a  $2\frac{1}{4} \times 3\frac{1}{4}$  Graflex cut film holder. The camera assembly is nickel plated and, to avoid reflections, the inner surface is blackened with black-board slating.

The choice of a lens is important because of the fine detail it must register and the many corrections involved in the manufacture. In this laboratory, micro tessar lenses are used which are corrected for spherical aberration, oblique aberration, axial chromatism, and lateral chromatism (3). These are 4 element lenses, especially designed for low power magnification, having extreme flatness of field, high speed, good image formation, and high resolving power. An  $f/4.5$  lens with a 48 mm. focal length has been found most useful for photographing grain; it covers a field of approximately  $1\frac{1}{2}'' \times 1\frac{1}{2}''$  and gives magnifications up to about 3 diameters at a lens-to-film distance of 192 mm. A second lens with a focal length of 72 mm. has also been used to obtain magnifications of lower diameters. For some work requiring greater magnification, say up to about 10 diameters, lenses of a focal length of 16 or 32 mm. may be required. A shorter focal length lens gives greater magnification at equal lens-to-film distances.

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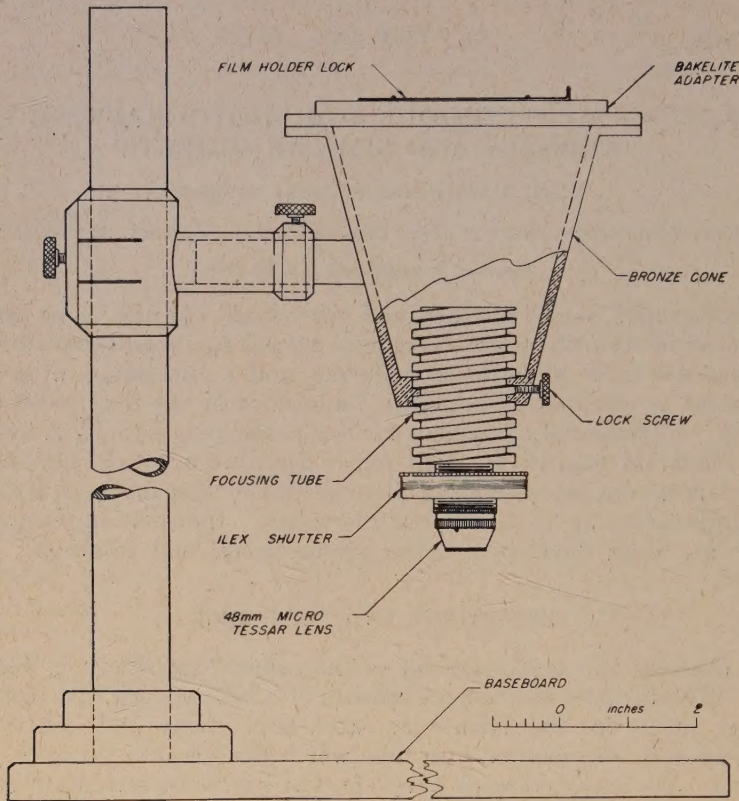


FIGURE 1. Drawing of Camera.

The camera has an Ilex Number 00 shutter with time, bulb, and speeds ranging from 1 second to 1/300 second.

A suitable stand for the camera is also shown in Figures 1 and 2. It provides means for raising or lowering the camera and for tilting it to any desired angle.

### *Lighting*

The detail in the photographs can be increased considerably by correct lighting. For the barley kernels shown in Figure 3 and for other specimens not requiring instantaneous shutter speeds, good results have been obtained with two 15-watt Cenco Universal microscope lamps. These occupy a minimum space and are easily adjusted for height and angle. The insect illustrated in Figure 4 is black and the only way to obtain variations in tone is to adjust the lighting correctly. To show up detail and bring up highlights, the angle of illumination should be about 20°, but this will vary considerably with the type of stage and the amount of detail to be shown. The artistic sense of the photographer is of first importance in this connection. A ground glass focusing panel is a great help in assessing the lighting, and it may be added that better results than those observed on the ground glass are rarely obtained in the finished photograph.





FIGURE 2. Photograph of Camera and Cenco Microscope Lamps.

If instantaneous shutter speeds are necessary, more concentrated and intense lighting is required. The photograph of live insects shown in Figure 5 was taken at a shutter speed of  $1/100$  second employing 3 number 1 photo-floods and a 150-watt reflector spot lamp for illumination. Control of lighting under these conditions is quite difficult. However, if powerful microscopic spot lamps and projection lamps are available many of these difficulties are minimized.

#### *Film*

Tri-X panchromatic film has been used for most of the work in this laboratory. It has an emulsion speed of 100 Weston rating, which makes it suitable for rapid shutter speeds. Ortho-X is also satisfactory and gives slightly higher contrasts. As a wider range of films becomes readily available, it may well be found that films of slower speed and finer grain are better for certain types of work.





FIGURE 3. Photograph of barley kernels showing detail obtained with the correct lighting, and using Edwal Super-20 for developing the films.

### *Exposure*

Experience in this laboratory suggests that the exposure time for black and white film is best determined by trial and error, as an exposure meter is difficult to use for close-up work. However, if conditions warrant the use of an exposure meter, it will be necessary to calculate the effective  $f$  value, which is decreased because the object is closer than eight times the focal length of the lens (4). The calculation may be made with the following formula (4):

$$\text{Effective } f \text{ value} = \frac{\text{Indicated } f \text{ value} \times \text{distance from lens to film}}{\text{Focal length of lens}}$$

Employing the two 15-watt Cenco microscope lamps and an indicated lens stop of  $f/16$ , exposure times of the order of 5 to 10 seconds have proved most satisfactory for grain kernels and similar objects.

### *Developer*

A developer which gives clean highlights and fine detail in the deepest shadows is required. Satisfactory results have been obtained with Edwal Super-20 fine grain developer (6) using 37 minutes developing time at a temperature of 70° F. Tests conducted at a wide range of exposure times indicated that this developer has considerable tolerance to variations in exposure. A modified DK-20 developer (2) has also been used. The developing time is shorter (16 min. at 70° F.) but the developer has less exposure tolerance and thus demands greater care in obtaining the correct time.





FIGURE 4. Yellow meal worm adult showing what can be done with suitable lighting.

#### *Enlarging*

As the engraver will prefer a larger photograph than is to be reproduced in the journal when making his cut, it is frequently necessary to make an enlargement unless a large camera and complete stock of lenses at various focal lengths are available. A condenser type enlarger fitted with a 72 mm. micro tessar lens is used in this laboratory. In most cases prints are made on Number 4 Glossy Kodabromide paper. Two developers, Edwal-102 (6) and Kodak D-72, have been used, and the results showed little difference between them. The developing time of Kodabromide paper in D-72 at 70° F. is from 1 minute to about 1½ minutes as compared with 2½ minutes to about 5 minutes with Edwal-102. The D-72 developer was chosen because of its shorter developing time.



FIGURE 5. Live Granary weevils photographed at instantaneous shutter speeds.



### *Reproduction*

The quality of paper that the cuts will be printed on is the determining factor for the type of cut to be made, and for the best results the advice of the photo engraver and the printer is invaluable. Illustrations of grain kernels must reproduce a high degree of detail. For those shown in Figure 3, a 150-line copper screen half-tone cut was made and printed on good quality dull enamel paper.

### *Colour Photography*

The equipment described has been used for colour photography, and some examples have been published elsewhere (5). Kodachrome professional type B film, having a colour temperature rating of 3200° K. (1, 4) and a film speed of 6 Weston units, was used. No. 1 photoflood lamps rated at a colour temperature of 3490° K. were employed for illumination. The truest colour rendition is obtained when the colour temperature of the lamps is 3200° K. However, a deviation of about 50° K. will give good transparencies. To obtain illumination of 3200° K. with the photofloods, the line voltage was reduced to 90 volts. The rule is that for a drop of 1 volt in the line, the colour temperature is reduced by approximately 10° K. Suitable filters are also available for increasing and decreasing the colour temperature of lamps, and a colour temperature meter is invaluable for determination of temperature.

Kodachrome has a very limited exposure latitude, and the exposure time must therefore be accurate. Moreover, as Kodachrome must be sent to the factory for developing, a test exposure cannot be made without considerable delay and expense. The Eastman Kodak Company recommend (4) that this difficulty be overcome by making a series of test exposures on Kodak Super-speed Direct Positive Paper. This is available in sheets or in film sizes, and processing instructions are enclosed with it. An alternate method, which has been used in this laboratory, is to make the test exposures on a black and white film with speed and exposure latitude closely similar to those of Kodachrome. Kodak 35 mm. Direct Positive movie film, developed in D-72 (1 part stock solution to 2 parts water) for five minutes at 70° F., is very satisfactory. Experience suggests that conditions equivalent to slight underexposure of the movie film give the best results with Kodachrome. However, in making the fine adjustments required to obtain the best transparency, individual judgment and preference play so large a part that specific advice can hardly be offered.

### SUMMARY

Apparatus and technique are described for photographing grain kernels, insects, and other specimens of similar size. The camera consists of a cast bronze cone with a lens mounted in a threaded tube for focusing on a ground glass. Good photographs of kernels and insects were obtained with Tri-X Panchromatic film developed in Edwal Super-20 developer. Correct lighting is important to obtain detail and variations in tone. For colour photography, colour temperature of the lamps and exposure time must be determined accurately.



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## MAXIMUM DEPTH OF SEEDING EIGHT CULTIVATED GRASSES<sup>1</sup>

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Seeding perennial forage crops at too great a depth is considered to be the principal cause of failure to secure satisfactory stands within the plains region of Western Canada. However, under these arid conditions, the deeper that seeds can be placed the better soil moisture may be for germination. Therefore, a desirable characteristic of any new species or variety would be ability to emerge from greater depths than commonly grown species or varieties. In addition, rapid rate of emergence also would be desirable, enabling plants to become well established while climatic conditions were favourable.

Recently, several relatively new grasses have shown promise of being suitable for much of this area. In an effort to supply data on agronomic characteristics a greenhouse experiment was conducted in 1946 to determine the maximum depth at which 4 of these grasses may be seeded and still obtain satisfactory emergence. Included in the experiment were 4 other standard grasses. This paper summarizes the results of the experiment.

### LITERATURE

Love and Hanson (5) obtained good emergence of crested wheat-grass at a  $\frac{3}{4}$ -inch depth of seeding on clay soil but at greater depths, emergence fell off sharply. Similar results are reported by Kirk, Stevenson, and Clarke (4).

Murphy and Arny (6) concluded, from field and greenhouse experiments, that depth of seeding was the most important factor governing seedling emergence. A  $\frac{1}{2}$ -inch depth was most satisfactory for the 10 crops studied on various soil types, although brome and Reed Canary grass gave satisfactory stands from greater depths. There were differences in the per cent emergence of the crops on different soil types. Environmental conditions influenced emergence considerably.

Clarke and Heinrichs (2) and Heinrichs (3) recommend shallow seeding for crested wheat-grass and sweet clover, not over 1 inch on clay soils and not over  $1\frac{1}{2}$  inches on sandy soils. Greenhouse experiments showed that better emergence of seedlings resulted on sandy loam soil than on clay loam at given depths. Although no brome seedlings came up when seeded 3 inches deep in clay loam soil, 53% emerged on sandy loam at this depth. Greater emergence resulted when the surface soil was dry than when it was moist.

Wasser and Nelson (7) state that satisfactory emergence of intermediate wheat-grass is obtained by seeding from  $\frac{1}{2}$  to  $1\frac{1}{2}$  inches deep and of Russian wild-rye grass from  $\frac{1}{2}$  to 1 inches.

<sup>1</sup> Contribution from the Division of Forage Plants, Experimental Farms Service, Dominion Department of Agriculture, Ottawa, Canada.

<sup>2</sup> Assistants in Forage Crops.



## MATERIALS AND METHOD

The following grasses were included in the experiment:

*Agropyron cristatum* (L.) Gaertn.  
*Agropyron trachycaulum* (Link.) Malte  
 var. *typicum* Fern.  
*Agropyron elongatum* (Host) Beauv.  
*Agropyron intermedium* (Host) Beauv.  
*Phalaris arundinacea* L.  
*Bromus inermis* Leyss.  
*Elymus junceus* Fisch.  
*Elymus virginicus* L. var. *submuticus*  
 Hook.

Fairway crested wheat-grass  
 Grazier slender wheat-grass  
 Tall wheat-grass  
 Ree intermediate wheat-grass  
 Reed Canary grass  
 Parkland brome  
 Russian wild-rye  
 Virginia wild-rye

Before seeding, germination tests were made on all seed lots. Adjustments in the amount of seed sown were made so that 100 theoretically viable seeds were sown in each plot. Haverhill loam soil, which had been air dried and sieved to remove lumps, was used for the experiment. Seeding was done in flats measuring 14 by 16 inches and 4 inches deep.

The grasses were sown at 6 depths,  $\frac{1}{2}$  inch, 1 inch,  $1\frac{1}{2}$  inches, 2 inches,  $2\frac{1}{2}$  inches and 3 inches. Each flat contained 6 plots of 2 rows each. One species was sown in each flat. This was accomplished by constructing wooden soil levellers of the required depths and planting the seed at the 6 depths consecutively, starting with the 3-inch depth. Six replicates

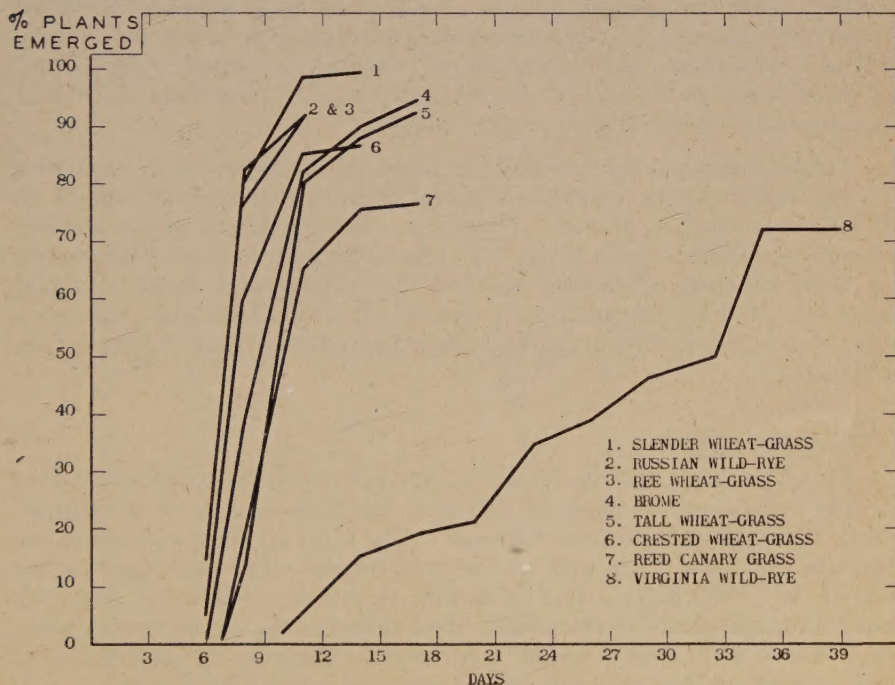


FIGURE 1. Rate of emergence of 8 grasses from  $\frac{1}{2}$ -inch depth.



were used, a replicate consisting of the 8 grasses each seeded at the 6 depths. The various depths of seeding were randomized within each flat and the flats were randomized within each replicate for position in the greenhouse.

Following seeding the soil was moistened thoroughly and kept at optimum moisture for the duration of the experiment. Notes were recorded daily on the number of days required for the emergence of plants from each depth. Total emergence counts were made every 3 days on all the plots. The experiment was terminated 39 days after seeding at which time all grasses, with the exception of Virginia wild-rye, had ceased to show any increase in total emergence.

#### RATE OF EMERGENCE

#### RESULTS

The rates of emergence of the 8 grasses from the  $\frac{1}{2}$ -inch depth (the optimum or near optimum depth for most of the species) are shown graphically in Figure 1. Five of the grasses began emerging 6 days after seeding while tall wheat-grass and Reed Canary grass emerged 1 day later and Virginia wild-rye 4 days later. Ree wheat-grass and Russian wild-rye were the first to complete emergence from the  $\frac{1}{2}$ -inch depth, 9 days after planting, followed by slender wheat-grass, crested wheat-grass, tall wheat-grass, brome and Reed Canary grass, the latter reaching maximum emergence 14 to 17 days after planting. The rate of emergence of Virginia wild-rye was very slow and was not complete at the end of the experiment.

The rate of emergence of each species from the various depths is shown in Figure 2. In general there is a lag of one day between depths but this increases with the greater depths. Emergence was complete from all depths in 20 days for crested wheat-grass and Russian wild-rye; in 23 days for Ree wheat-grass; in 26 days for tall wheat-grass, slender wheat-grass and brome; and in 29 days for Reed Canary grass. Virginia wild-rye had not completely emerged 39 days after planting.

The slow rate of emergence of Virginia wild-rye appeared to be related to some extent to the greenhouse temperature which varied from 65 to 90° F. between night and day. There was also variation in temperature in different parts of the greenhouse. In the cooler end very poor emergence was obtained from all depths while at the warmer end there was good emergence from all depths up to 2 inches. It would seem that this grass requires a higher temperature to induce germination than others in the experiment.

#### TOTAL EMERGENCE

The experiment was designed so that individual analysis of the data for each grass could be made as well as a complete analysis of the experiment. Both procedures were followed. The data on total emergence are presented in Table 1 as well as the transformed data according to the formula  $P = \sin^2 \theta$  (1). The minimum significant difference given for each crop applies only to the transformed data as does the minimum significant difference for the whole experiment. Because the emergence of Virginia wild-rye was not complete, it was analyzed by itself only.



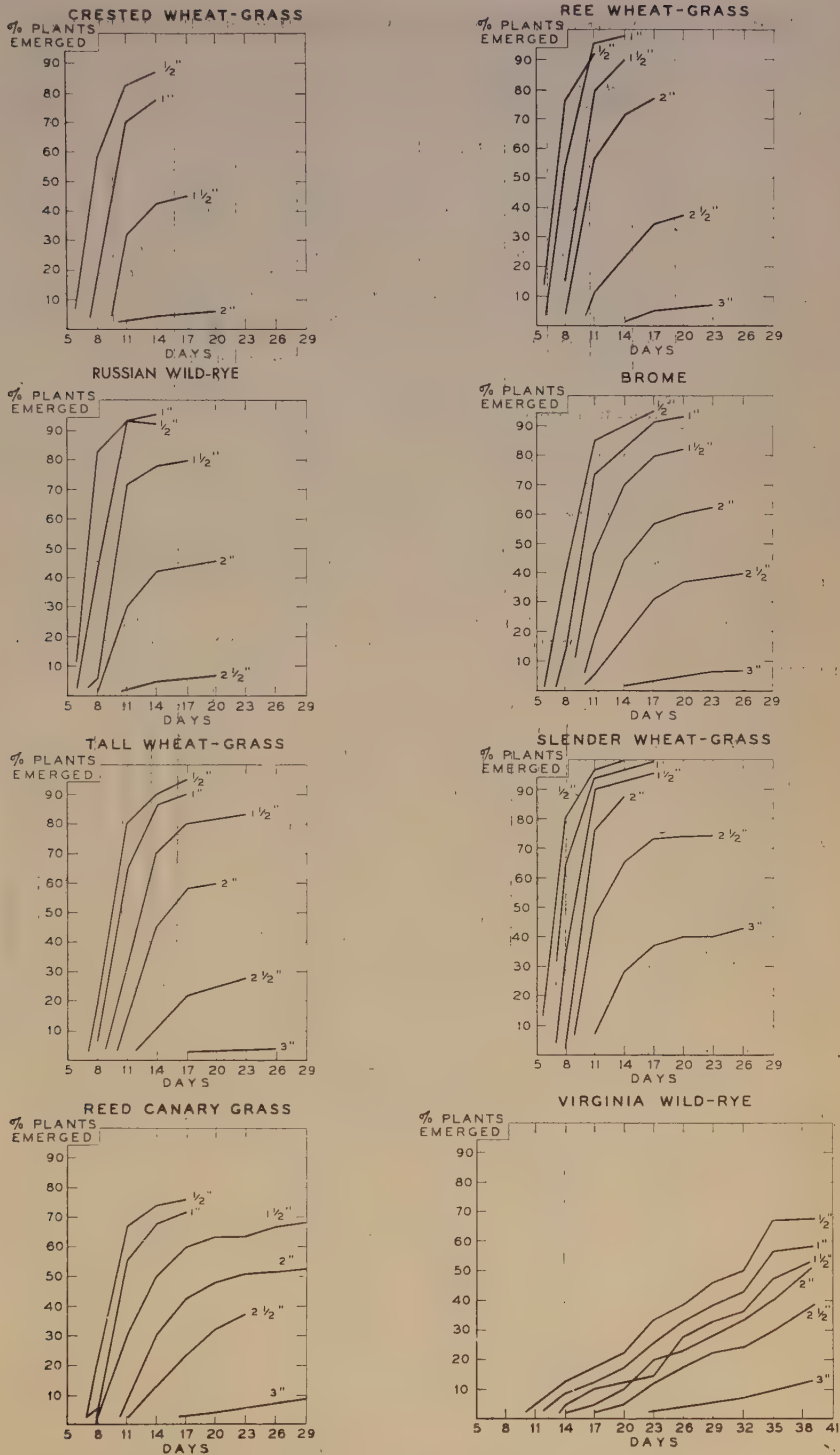


FIGURE 2. Rate of emergence of the 8 grasses when seeded at different depths ( $\frac{1}{2}$ , 1,  $1\frac{1}{2}$ , 2,  $2\frac{1}{2}$ , and 3 inches).



TABLE 1.—TOTAL EMERGENCE OF 8 GRASSES FROM 6 DEPTHS OF SEEDING

Species	Average % emergence from different depths in ins.						Average of data converted to $\sin^2 \theta$						Signi- ficant differ- ence
	$\frac{1}{2}$	1	$1\frac{1}{2}$	2	$2\frac{1}{2}$	3	$\frac{1}{2}$	1	$1\frac{1}{2}$	2	$2\frac{1}{2}$	3	
Virginia wild-rye	66	58	53	50	39	11	56.9	49.9	46.5	44.1	37.0	14.4	11.4
Crested wheat-grass	87	79	44	6	0	0	69.6	63.6	41.6	12.6	0	0	5.98
Slender wheat-grass	99	99	97	89	74	41	87.4	86.7	83.2	72.9	59.6	40.1	6.66
Tall wheat-grass	93	90	83	61	27	3	75.8	72.6	66.4	51.6	31.0	8.9	7.47
Ree wheat-grass	92	98	90	77	38	6	75.7	86.3	71.4	61.4	37.9	13.2	8.43
Brome	94	94	83	62	40	8	76.9	76.4	65.6	51.9	38.7	13.5	8.97
Reed Canary grass	76	73	67	54	37	9	60.8	58.7	54.8	47.1	37.4	15.3	6.35
Russian wild-rye	93	94	80	46	8	0	76.8	77.2	64.1	42.5	15.7	1.4	5.67

Minimum significant difference for entire experiment (7 species) = 7.11

The F values obtained in the analyses of the data were highly significant for depths, species and the interaction of depths and species. The standard error of the experiment was 4.94%.

The total emergence of all species from the various depths at the end of the experiment is shown graphically in Figure 3.

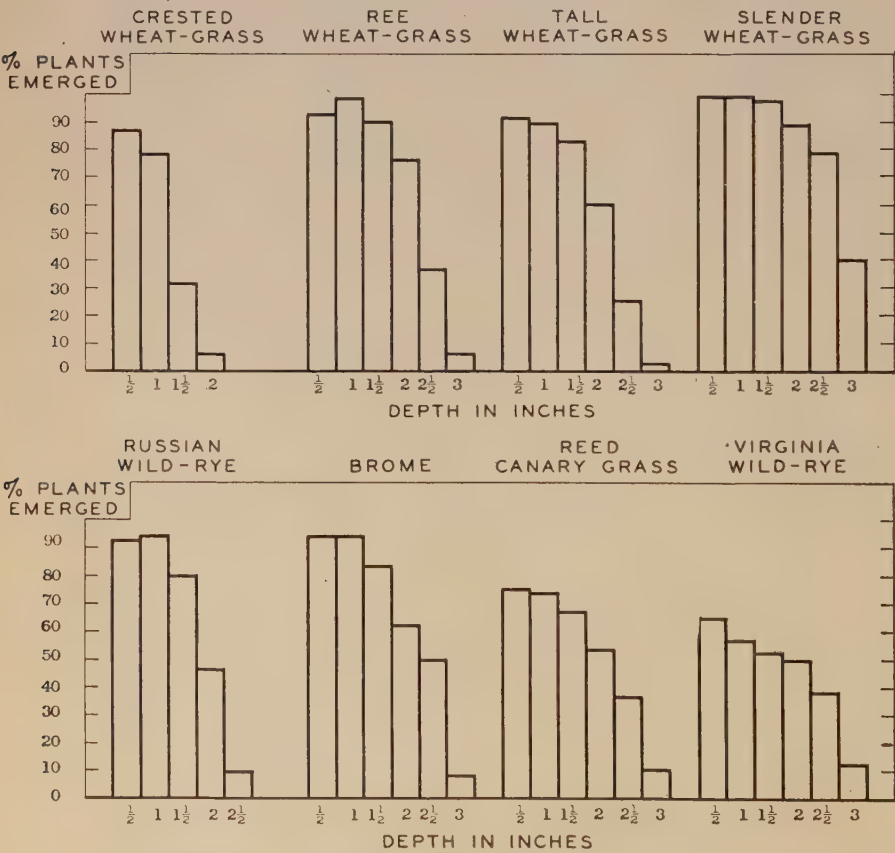


FIGURE 3. Total emergence of 8 grasses from 6 different depths. (Virginia wildrye still increasing 39 days after planting).



## DISCUSSION

The data indicate that under the conditions of this experiment the maximum depth at which the species studied may be sown on loam soil is as follows:

Crested wheat-grass	1 inch
Russian wild-rye	1½ inches
Brome	1½ inches
Tall wheat-grass	1½ inches
Ree wheat-grass	2 inches
Reed Canary grass	2 inches
Virginia wild-rye	2 inches
Slender wheat-grass	2½ inches

It may be stated further that tall wheat-grass, Ree wheat-grass, Russian wild-rye and Virginia wild-rye can be seeded deeper than crested wheat-grass and as deep or deeper than brome. This is a favourable factor when establishing stands within low rainfall regions. However, the slow rate of emergence of Virginia wild-rye is a distinct disadvantage.

It is of interest to note the good emergence of Reed Canary grass, a small seeded grass, from the 2-inch depth and that of slender wheat-grass from a 2½-inch depth.

## SUMMARY

A greenhouse experiment was conducted on maximum seeding depth with 4 promising grasses, Russian wild-rye, Virginia wild-rye, tall wheat-grass and Ree wheat-grass in comparison with 4 standard grasses, crested wheat-grass, slender wheat-grass, brome and Reed Canary grass. All species, with the exception of Virginia wild-rye, emerged rapidly from the optimum depth of ½ inch and emergence was complete in from 9 to 17 days after seeding. The total emergence of the eight species from the 6 depths of seeding (½, 1, 1½, 2, 2½ and 3 inches) on Haverhill loam soil indicated that the maximum depth at which the 4 new species could be seeded was greater than that for crested wheat-grass (1 inch) and equal or greater than for brome (1½ inches). The slow rate of emergence of Virginia wild-rye would discriminate against it as a suitable grass for low rainfall areas but Russian wild-rye, Ree wheat-grass and tall wheat-grass appear to be equal or superior to standard grasses in this respect.

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# FEEDING TRIALS WITH CHICKS OF A NEW VITAMIN A AND D CARRIER FOR POULTRY<sup>1</sup>

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One of the operations most unpopular with the manufacturers of poultry and swine feeds is the handling and mixing of the vitamin-bearing oils, which constitute the only non-solid ingredient of most formulae. The destruction in mixed feeds of the vitamins, particularly A, from the common oil sources is a rather serious problem.

There are available a number of solid preparations supplying vitamin D, but few, if any, solid carriers of both vitamins A and D have been available in Canada. In view of the impossibility of depending upon yellow corn and sun-cured or dehydrated green feeds for the vitamin A activity in Canadian poultry feeds, supplementary sources of vitamin A are essential for such rations.

Recently, a solid carrier of both vitamins has become available. This product, referred to hereinafter as "A and D meal," of Canadian origin<sup>4</sup> is described as a "preparation of vitamins A and D in Dry Herring Meal Base." An advantage claimed for it, in addition to greater convenience in handling and mixing, is a greater stability of the vitamin content, particularly after mixing with rations, than is found with the customary oil sources of these vitamins. This is achieved by the use of stabilizing agents in the product. Tests of this material in rations for starting and growing chicks were conducted on a practical basis, in an attempt to evaluate the acceptability of the product for such rations.

## EXPERIMENT 1

The first test was planned as an evaluation on a practical basis of the acceptability of "A and D Meal" as a substitute for a feeding oil in a ration for growing chicks, judged chiefly on the basis of growth.

## EXPERIMENTAL

Eight groups of 36 newly-hatched Barred Plymouth Rock chicks with uniform sex distribution were placed in similar compartments of electrically-heated battery brooders maintained in the air-conditioned biological laboratory. At the end of 5 weeks, the birds were moved to growing batteries in the same laboratory, and were maintained in these until they reached an age of 10 weeks, at which time the experiment was concluded. Feed and water were before the birds at all times.

The birds in Groups 1 to 4 received Ration No. 1, and those in Groups 5 to 8 received Ration No. 2. The composition of these rations is shown in Table 1. It will be noted that the two rations were identical except for the supplementary sources of vitamins A and D. In Ration No. 1, these

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<sup>4</sup> A product of the Canadian Fishing Co., Ltd., Vancouver, B.C.



vitamins were supplied by 0.15% of a poultry feeding oil guaranteed to contain 400 A. O. A. C. units of vitamin D and 3000 International units of vitamin A per gram. In Ration No. 2, the same amounts of vitamins A and D were furnished by 0.30% of the "A and D Meal," which was guaranteed to contain 200 A. O. A. C. units of vitamin D and 1500 International units of vitamin A per gram. The amounts of these vitamins supplied in the two rations are somewhat less than commonly recommended for inclusion in practical rations although they are slightly in excess of published allowances. The purpose in avoiding any great over-supply or margin of safety in these rations was to avoid masking any practical loss or inadequacy in either vitamin supplement.

TABLE 1

Ingredient	Weight (pounds)	
	Ration No. 1	Ration No. 2
Ground yellow corn	10.0	10.0
Ground barley	14.35	14.2
Wheat bran	10.0	10.0
Wheat shorts	10.0	10.0
Ground wheat	12.0	12.0
Ground whole oats	10.0	10.0
Rolled oat groats	10.0	10.0
Dehydrated alfalfa	2.0	2.0
Cereal grass	1.0	1.0
Soybean oil meal	7.5	7.5
Meat meal	3.0	3.0
Fish meal	5.0	5.0
Buttermilk powder	1.25	1.25
Oyster shell	2.25	2.25
Insoluble grit	1.0	1.0
Iodized salt	0.5	0.5
"A and D Meal"*	—	0.3
Fortified fish oil**	0.15	—
Total	100.0	100.0
Protein (%)	19.3	19.4
Calcium (%)	1.7	1.7
Phosphorus (%)	0.8	0.8

\* 1500A, 200D. \*\* 3000A, 400D.

To each ration was added:

Crystalline riboflavin at the rate of 1.28 grams per ton.

Manganous sulphate ( $\text{MnSO}_4 \cdot 4\text{H}_2\text{O}$ ) at a rate of 4 ounces per ton.

In addition to the grit contained in the ration insoluble grit was supplied *ad libitum* to all chicks from the age of five weeks.

These rations were mixed in 500 pound lots at frequent intervals throughout the experimental period. Of each lot, one-half was fed and the other half stored for use in the stability test (Experiment 2).

All birds were weighed at the end of 4, 6, 8 and 10 weeks. In addition, records were kept of mortality and the incidence of any abnormalities, and the birds were observed for feathering and general appearance.

The birds were sexed again at the end of 10 weeks to correct any errors in the initial sex records.



## RESULTS

The weight and mortality records are summarized in Table 2.

Application of the conventional methods of variance analysis to the weight data revealed no significant differences in weight attributable to diet at any of the weighings. Mortality was quite low on both rations and there was no apparent ration effect in the few cases of perosis, the only abnormality which was noted in any of the birds. No differences in feathering or general appearance of the birds could be detected.

It is concluded that, with the type of ration used, the "A and D Meal" was a quite satisfactory substitute for the vitamin-bearing poultry oil for chicks up to 10 weeks of age as judged by weight, feathering and general appearance of the birds.

TABLE 2.—WEIGHT AND MORTALITY DATA, EXPERIMENT 1

Ration No.	Group No.	Orig. No. each sex		Average weight (grams)				Total* mortality (all causes)	Lbs. feed per lb. gain
				4 weeks	6 weeks	8 weeks	10 weeks		
1	1	M	15	188	370	672	1097	1	4.37
(Fortified fish oil)		F	21	213	397	670	994		
	2	M	18	219	417	722	1138	1	
		F	18	224	421	696	1032		
	3	M	20	206	401	698	1087	3	
		F	16	242	447	711	1043		
	4	M	17	213	416	716	1114	2	
		F	19	227	407	690	1023		
2	5	M	17	209	415	724	1150	2	4.56
(“A and D Meal”)		F	19	222	420	693	1025		
	6	M	16	202	406	727	1177	4	
		F	20	219	409	695	1020		
	7	M	16	199	406	743	1143	3	
		F	20	233	433	709	1055		
	8	M	16	209	403	721	1188	3	
		F	20	214	390	657	1014		

\* Includes those birds which, having developed severe perosis, were destroyed at 8 weeks. This involved 4 birds on Ration 1 and 5 birds on Ration 2.

## Experiment 2

This experiment was designed to compare the ability of two chick rations, one containing vitamins A and D in the form of a fortified fish oil and the other containing "A and D Meal," to support growth in chicks after storage under practical conditions. In addition a further test of the aspect studied in Experiment 1 was included.

## EXPERIMENTAL

Sixteen groups of 37 newly-hatched Barred Plymouth Rock chicks were placed in pens of electrically-heated battery brooders. Sex distribution was uniform throughout the groups. The pens, management, and general routine of the experiment were similar to those in Experiment 1.

Four different rations were fed. The birds in Groups 1 to 4 received Ration No. 1 (old), those in Groups 5 to 8 received Ration No. 2 (old), those in Groups 9 to 12 received Ration No. 1 (new) and those in Groups 13 to 16 received Ration No. 2 (new).

The two "old" rations were stored portions of the complete rations mixed during the period of Experiment 1, Ration No. 1 containing fortified fish oil and Ration No. 2 containing "A and D Meal." These rations had been stored at the normal room temperature of an unheated and uninsulated storage building for a period of approximately 7 months. The period of storage for the earliest-mixed lot was from July to February and for the latest-mixed lot from September to April. In order that the storage periods might be approximately the same for all lots of the "old" rations, the various lots were fed in the order in which they had been prepared.

The two "new" rations were mixed as required during the experimental period, the composition of the two rations being identical with that of the "old" Ration Nos. 1 and 2, respectively. The 200D, 1500A "A and D Meal" used in the "new" Ration No. 2 was of recent preparation.

All birds were weighed at the end of 4, 7, and 10 weeks, and judged as before.

## RESULTS

The weight and mortality data are presented in Table 3.

TABLE 3.—WEIGHT AND MORTALITY DATA, EXPERIMENT 2

Ration No.	Group No.	Orig. No. each sex		Average weight (grams)			Total mortality* (all causes)	Lbs. feed per lb. gain
				4 weeks	7 weeks	10 weeks		
1	1	M	22	279	648	1221	2	3.2
Old (fish oil)	2	F	15	258	542	913		
		M	22	268	646	1231	5	
		F	15	263	556	913		
3	M	20	252	626	1248	2		
		F	17	265	566	1037		
4	M	19	248	602	1234	4		
		F	18	239	528	1010		
2	5	M	18	265	618	1186	2	3.7
Old ("A and D Meal")	6	F	19	257	528	969		
		M	12	283	681	1255	2	
		F	25	262	583	1046		
7	M	16	272	634	1206	1		
		F	21	234	541	1019		
8	M	16	311	645	1284	3		
		F	21	235	558	1071		
1	9	M	14	311	723	1301	2	3.6
New (fish oil)	10	F	23	286	606	1068		
		M	21	289	694	1282	3	
		F	16	272	616	1073		
11	M	15	304	720	1267	2		
		F	22	276	609	1035		
12	M	19	314	694	1271	0		
		F	18	289	611	1090		

\* Includes those birds which were destroyed following the development of severe perosis. This involved 2 birds on "old" Ration 2, 1 bird on "new" Ration 1, and 2 birds on "new" Ration 2.



TABLE 3.—WEIGHT AND MORTALITY DATA, EXPERIMENT 2—*Concluded*

Ration No.	Group No.	Orig. No. each sex		Average weight (grams)			Total mortality* (all causes)	Lbs. feed per lb. gain
				4 weeks	7 weeks	10 weeks		
2	13	M	17	300	740	1323	2	3.6
New ("A and D Meal")		F	20	266	579	1006		
	14	M	19	311	712	1305	3	
		F	18	272	595	1070		
	15	M	21	294	681	1272	1	
		F	16	262	589	1074		
	16	M	20	312	720	1272	2	
		F	17	301	682	1117		

\* Includes those birds which were destroyed following the development of severe perosis. This involved 2 birds on "old" Ration 2, 1 bird on "new" Ration 1, and 2 birds on "new" Ration 2.

The results indicate that there was no appreciable difference between the groups fed the two "new" rations, nor between the groups fed the two "old" rations. Variance analysis of the weight data revealed no significant differences between the weights resulting from the two "new" rations or between the weights of birds receiving the two "old" rations. No differences in feathering or appearance of the birds were apparent, and no marked ration influence is evident in the mortality data.

It is concluded that Ration Nos. 1 and 2 underwent storage equally well as judged by the growth of chicks to 10 weeks.

It will be noted that the weights of birds receiving the "old" rations were lower throughout than those of birds fed the "new" rations. These differences in weight were found to be highly significant at 4 and 7 weeks, but not significant at 10 weeks. Whether any particular importance can be attached to this last observation it is impossible to say. In this connection it is interesting to note that the feed consumption was lower on both the "old" rations, particularly "old" Ration No. 1. Only in the latter did the feed:gain ratio differ from that on the "new" rations, but it is impossible to say whether this slight difference is of significance.

The observation that weight gains were less on the stored rations than on freshly-mixed rations is a common one. The present study gives no indication as to which factor or factors in the ration may be associated with the decreased efficacy of the ration.

Further studies to compare calcification in chicks receiving the two types of supplement are contemplated.

#### SUMMARY

Two experiments, in which, the weight of chicks to 10 weeks of age has been used as the chief criterion, have been conducted to compare the value in a practical ration of a new source of "vitamins A and D in Dry Herring Meal Base" with that of a conventional fortified fish liver oil. Under the conditions of the experiment, the new product was found to be a satisfactory substitute for the oil.

In a test of mixed rations containing the different sources of vitamins A and D the weights of chicks up to 10 weeks of age indicated no practical differences in stability of the vitamins from the two sources.

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# THE RESISTANCE OF WHEAT VARIETIES TO SEED BLEACHING<sup>1</sup>

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## INTRODUCTION

The bleaching or weathering of ripe grain, standing uncut in the field awaiting the combine, or harvested grain in the swath or stook awaiting the combine or separator, results in lowered grades and economic loss to the grower. These losses are most frequent and severe in areas subject to spells of wet harvest weather. Furthermore, some varieties are known to bleach or weather much more readily than others.

Since a variety which retains its seed color is of more commercial value than one which does not, other properties being equal, an investigation was carried on in 1944 and 1945 to determine the degree of resistance to seed bleaching of wheat varieties and hybrid lines at University Farm, Saskatoon.

## LITERATURE

The literature on seed bleaching of wheat is not extensive and is particularly deficient as to tests of difference between varieties.

Whitcomb and Johnson (1) investigated the effects of severe weathering on wheat and found no severe disturbance of the quality of the grain. Bracken and Bailey (2) studied the effect of delayed harvesting on the quality of wheat. They concluded that "dark, hard wheat of the Turkey Red type does not deteriorate in quality upon standing uncut in the field when subjected to alternate wetting and drying in spite of the fact that the grains bleach and lose weight per measured volume." Jewell and Miller (3) in Australia concluded that, apart from lowered bushel weight and bleached appearance, exposure of ripe grain in the ear to heavy rain followed by drying before harvest has no appreciable deleterious effect on the flour yield and baking qualities. Cayzar (4) found that light bleaching had no noticeable effect upon the milling or baking quality of Australian wheats but that severe bleaching actually tended to improve the milling and baking qualities of hard wheats, the degree of improvement depending upon the variety. Atkinson (5) found that bleached wheat had a lower bushel weight on account of an increase in volume due to the alternate wetting and drying. Copp (6) exposed 6 lines of wheat, under field conditions to artificial and natural rainfall alternated with fair drying conditions, for a period of 3 weeks. This heavy weathering did not result in any significant losses in grain weight or yield.

## MATERIALS AND METHODS

In 1944 head samples were taken from every plot of the replicated yield tests of spring wheat varieties and purified hybrid lines at University Farm, Saskatoon. Similar samples were taken in 1945 from varieties and

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lines which had been grown in the nursery in 1944 and again in 1945, thus permitting a 2 year study of the same varieties. Each sample was taken from a border row of each plot immediately upon its' maturity and divided into 2 lots, designated as I and II, of approximately 22 heads per lot.



FIGURE 1. Lot I head samples from individual plots being exposed to the weather after the removal of the paper covering.

Each unthreshed sample of Lot I was wrapped in brown paper to protect it from the weather and tied to a stake, driven into the ground at the plot location as shown in Figure 1. Two days after the last plot of each particular test had ripened the paper was removed, thus exposing all the samples in each test to the same set of weather conditions at the same time. This provided a uniform test of both early and late maturing varieties. Three days after the Lot I heads were exposed to the weather one-quarter of them were taken into the laboratory where they were threshed and scored for seed color. Similar samples were taken and scored 10, 17 and 24 days after the start of exposure.

The Lot II samples were exposed to the weather as threshed grain. Before exposure each sample was divided into 3 parts, known as A, B, and C. The A samples were exposed to the weather on wooden trays in the field, the trays being protected from birds and gophers by means of a wire cage as shown in Figure 2. An estimate of the color of each sample was made on the 1, 2, 3, 6, 10, 15 and 20 day of exposure. The B samples of Lot II were exposed and scored in a similar manner as soon as the scoring of the A samples was completed. The C samples of Lot II were reserved for a bleaching test using a laboratory method.

In addition to the foregoing material a further set of samples, known as Lot III was taken 2 weeks after the last plot of each test had ripened. These samples were threshed and scored for seed color in the laboratory.



FIGURE 2. Wire cage used to protect the samples of Lot IIA and Lot IIB from birds and gophers.

All scoring was done by visually estimating the color score of each sample using as a guide both weathered and unweathered samples of standard wheat varieties. All varieties and lines used were also scored for color at maturity prior to being exposed to the weather. A variety was considered to be mature when the kernels could be indented with difficulty by the thumbnail. A record of the precipitation, temperature (minimum and maximum), hours of sunshine and wind velocity was kept for each day that the varieties were exposed.

Comprehensive laboratory tests were made to simulate the effects produced by natural weathering of unthreshed heads. Five groups of tests on 10 varieties and lines were made as follows: (A) The 10 samples were immersed in tap water at 55° F. in petri dishes for 10 minutes and then dried in a forage dryer operated at 84° F. The procedure was then repeated on fresh samples of the 10 varieties using drying temperatures of 175° F., 200° F. and 270° F. The drying time required was approximately 30 minutes. The samples were scored for color immediately after treatment. (B) Three sets of samples were immersed in tap water at 55° F. for periods of 10, 30 and 75 minutes, respectively, then dried at 200° F. in the forage dryer and scored for color. (C) Four sets of samples were immersed for 15 minutes in tap water at 55° F. and then air dried at (a) a temperature of 70° F. with the humidity at 25%; (b) 70° F., humidity 30%; (c) 70° F. and 70%; (d) 75° F., and 95%. The samples were scored when dry. (D) Three sets of samples were immersed in tap water at temperatures of 55° F. and 180° F. for 10 minutes and 212° F. for 3 minutes then dried at 200° F. and scored. (E) One set of samples was immersed in tap water at 55° F. for 10 minutes and dried at 200° F. in the forage drier. This procedure was carried out 4 times on the same samples. Color scores were taken after each treatment. Duplicate samples were used in all tests.



All Lot II C samples of both 1944 and 1945 were tested for retention of seed color by immersing them in tap water at 55° F. for 10 minutes and drying them in the forage dryer at 200° F.

TABLE 1.—SUMMARIZED 1945 RESULTS ON THE RETENTION OF SEED COLOR OF THE VARIETIES OF TEST V WHEN EXPOSED TO THE WEATHER, STANDING UNTHRESHED SUBSEQUENT TO THEIR MATURITY, FOR 4 DIFFERENT PERIODS OF TIME. (Lot I)

Variety	0 Days	3 Days score*	10 Days score	17 Days score	24 Days score	Average omitting 0 days
XM-4	94	91	82	71	67	77.8
Huron	92	92	80	71	67	77.5
XM-76	93	89	78	68	66	75.2
CA-11	89	88	74	66	61	72.3
MA-261	94	86	77	65	61	72.2
TA418-1	93	86	76	64	61	71.8
MA131-1	92	88	76	64	58	71.5
TA399-1	93	85	76	63	59	70.8
TA185-6	91	84	74	62	60	70.0
Henry	89	86	71	63	60	70.0
MA-163	94	85	74	63	56	69.5
TA526-2 × Ap	92	83	73	62	56	68.5
Apex	92	84	74	59	53	67.5
TA357	93	81	73	59	56	67.2
TA622-3	88	82	73	59	55	67.2
CA-6	92	86	74	57	54	67.8
TA998-4	88	81	71	59	56	66.8
XM76-1 × Th	91	75	72	61	59	66.8
TA700-1 × Ap	89	80	70	59	56	66.2
TA1001-2	86	78	69	57	53	64.2
TA622-2 × Ap	89	74	70	59	54	64.2
TA296-3	91	80	67	55	52	63.5
Regent	82	71	63	57	52	60.8
Thatcher	83	76	65	52	46	59.8
Sig. diff. at 5% level		6.4	5.8	5.2	6.1	

\* Each score is the mean of six replicates.

Abbreviations: XM—(sib Apex-41 × sib Apex-660) × Marquis; CA—Comet × Apex; MA—Marquis × Apex; TA—Thatcher × Apex; Th—Thatcher; Ap—Apex.

#### VARIANCE ANALYSIS (after 24 days of exposure)

Source of error	Sum of squares	Degrees of freedom	Mean square	F value	5% point	1% point
Total	7578	143				
Varieties	3331	23	144.8	4.09	1.60	1.94
Error	4247	120	35.4			

Mean = 57.4

SE<sub>v</sub> = 2.43

Necessary difference = 6.07

SE<sub>v</sub>% = 4.23

#### RESULTS

In the 2 years 1944 and 1945, a total of 152 varieties and hybrid lines of wheat were tested for their retention of seed color. Owing to the large amount of data only the results of Test V in 1945 will be given in detail. There results are shown in Tables 1 to 5, inclusive.

The results on Lot I of Test V are given in Table 1 the varieties being arranged in descending order of their average scores. Statistical analysis was applied to the results from each date on which color scores were taken. Significant differences were found to exist between the varieties and lines tested. The significant difference calculated for the results of each date divided the varieties into 3 levels of resistance. Huron, XM-4, XM-76 and MA-261 showed high resistance, while Thatcher, Regent and TA1001-2 showed low resistance to seed bleaching. The remaining varieties were more or less intermediate in their retention of seed color. Although the varieties lost color progressively as the time of exposure increased from 3 to 24 days, their relationships were not significantly changed. For example, XM-4 and Huron remained high while Thatcher and Regent remained low. This was further substantiated by calculating interactions using all possible combinations of results at the four scoring dates. In no case were the interactions significant at either the 5% or the 1% level as shown by the first six interactions given in Table 5.

From the close agreement between the results of each of the four scoring dates it is apparent that even the 3 and 10 day results are a reliable indication of the ability of a variety to resist seed bleaching. It is also of interest to note that after 17 days exposure the varieties tended to lose color at a much slower rate.

TABLE 2.—SUMMARIZED 1945 RESULTS ON THE RETENTION OF SEED COLOR OF THRESHED GRAIN OF THE VARIETIES OF TEST V WHEN EXPOSED TO THE WEATHER FOR 7 DIFFERENT PERIODS OF TIME SUBSEQUENT TO MATURITY. (LOT IIA)

Variety	0 Days score	1 Day score*	2 Days score	3 Days score	6 Days score	10 Days score	15 Days score	20 Days score	Average omitting 0 days
XM-4**	94	94	94	92	85	77	76	73	84.4
XM-76	93	92	92	89	84	78	76	70	83.0
Huron	92	91	90	88	85	78	77	71	82.9
MA-261	94	94	93	91	83	76	72	70	82.7
TA418-1	93	93	92	90	81	75	74	69	82.0
TA399-1	93	93	92	89	81	73	71	66	80.7
MA-163	94	93	92	89	80	72	69	66	80.1
MA131-1	92	91	90	87	80	74	71	67	80.0
TA357	93	92	91	89	81	72	69	65	79.9
TA185-6	91	91	91	88	80	72	70	66	79.7
XM76-1 × Th	91	90	90	87	79	72	69	66	79.0
Henry	89	89	88	86	81	73	70	64	78.7
CA-11	89	88	88	85	79	72	69	66	78.1
TA526-2 × Ap	92	92	91	88	79	69	66	62	78.1
TA622-2 × Ap	89	89	88	86	79	70	67	66	77.9
CA-6	92	91	90	87	77	70	64	62	77.3
Apex	92	91	90	86	76	68	65	63	77.0
TA700-1 × Ap	89	89	88	85	77	69	65	61	76.3
TA998-4	88	88	87	85	76	68	65	60	75.6
TA296-3	91	90	88	84	77	67	63	60	75.6
TA1001-2	86	86	85	82	77	69	66	63	75.4
TA622-3	88	87	87	84	76	68	63	59	74.8
Regent	82	81	81	79	76	68	65	60	72.9
Thatcher	83	82	81	80	72	62	58	55	70.0
Sig. diff. at 5% level						4.6		4.8	

\* Each score is the mean of six replicates.

\*\* See list of abbreviations given under Table I.



The results of Lot IIA and Lot IIB are presented in Tables 2 and 3, respectively. The varieties are arranged in descending order of average score. The observations from the 10 and 20 day exposures showed statistically significant differences between varieties. The significant difference calculated for each of the two dates of each test again showed XM-4, Huron, XM-76 and MA-261 to possess high resistance, while Thatcher, Regent and TA1001-2 showed low resistance to bleaching. The interactions between (a) the 10 and 20 day exposures of Lot IIA, (b) the 10 and 20 day exposures of Lot IIB, (c) the 10 day exposure of Lots IIA and IIB and (d) the 10 day exposure of Lots IIA and I all lacked significance. Thus the varieties reacted in a similar manner when exposed to the weather whether as unthreshed heads or as threshed grain.

TABLE 3.—SUMMARIZED 1945 RESULTS ON THE RETENTION OF SEED COLOR OF THRESHED SEED OF THE VARIETIES OF TEST V WHEN EXPOSED TO THE WEATHER FOR 7 DIFFERENT PERIODS OF TIME SUBSEQUENT TO MATURITY. (Lot IIB)

Variety	0 Days score	1 Day score*	2 Days score	3 Days score	6 Days score	10 Days score	15 Days score	20 Days score	Average omitting 0 days
XM-4**	94	90	73	72	71	70	66	64	72.3
Huron	92	88	71	71	69	69	64	61	70.4
XM-76	93	88	71	70	69	69	64	60	70.1
MA-261	94	89	70	70	68	67	62	60	69.4
TA399-1	93	89	69	68	66	65	59	57	67.6
TA418-1	93	87	70	69	66	64	59	56	67.3
MA-163	94	88	68	67	65	63	59	57	66.7
TA-357	93	86	69	67	65	64	59	56	66.6
CA-6	92	86	67	67	66	65	59	55	66.4
TA185-6	91	86	69	67	65	63	58	55	66.1
CA-11	89	85	68	67	65	64	58	54	65.9
MA131-1	92	86	67	67	65	63	58	55	65.9
Henry	89	85	68	67	65	63	57	55	65.7
XM76-1 × Th	91	85	67	67	65	63	57	53	65.3
TA526-2 × Ap	92	86	68	67	64	62	57	54	65.4
TA622-2 × Ap	89	86	68	67	64	63	56	54	65.4
Apex	92	87	66	66	63	60	55	52	64.1
TA998-4	88	85	67	65	63	61	55	53	64.1
TA700-1 × Ap	89	85	66	64	61	59	54	52	63.0
TA296-3	91	83	65	64	61	59	53	50	62.1
TA622-3	88	82	64	63	61	59	52	50	61.6
Regent	82	79	63	62	60	58	54	50	60.9
TA1001-2	86	81	64	62	59	59	53	48	60.9
Thatcher	83	79	60	58	56	54	49	46	57.4
Sig. diff. at 5% level						4.5		5.3	

\* Each score is the mean of six replicates.

\*\* See list of abbreviations given under Table 1.

Bleaching appeared to be caused chiefly by alternate wetting and drying. Exposure to sun, wind and possibly slight dew brought about some bleaching, as shown by Table 2, when rain did not occur until the fourth day of exposure. On the other hand, less than  $\frac{1}{10}$  inch of rain on the first day of exposure caused a striking loss of color in all of the samples of Lot II B (Table 3).

Table 4 gives the results of Lot IIC and Lot III as compared with the results of Lots I, IIA and IIB after 10 days exposure. Very close agreement between the results of the 5 treatments is apparent from the data. Statistical analyses of the results on Lot IIC and Lot III, as well as the interactions calculated between Lot IIC and Lots I, IIA and IIB, respectively, and between Lot I and Lot III confirm this close agreement. The results of the above interactions are given in Table 5.

TABLE 4.—COMPARISON OF THE RESULTS FROM 5 DIFFERENT METHODS OF TESTING THE VARIETIES AND HYBRID LINES OF TEST V FOR RETENTION OF SEED COLOR

Variety	0 Date	Lot I*	Lot IIA	Lot IIB	Lab. Dryer Lot IIC	Lot III
XM-4**	94	82	77	70	73	66
Huron	92	80	78	69	71	66
XM-76	93	78	78	69	71	62
CA-11	89	74	72	64	67	60
MA-261	94	77	76	67	68	61
TA-418-1	93	76	75	64	65	59
MA-131-1	92	76	74	63	67	57
TA-399-1	93	76	73	65	67	57
TA-185-6	91	74	72	63	67	57
MA-163	94	74	72	63	66	57
Henry	89	71	73	63	67	57
TA-526-2 × Ap	92	73	69	62	63	55
TA-357	93	73	72	64	64	55
Apex	92	74	68	60	65	55
TA-998-4	88	71	68	60	63	54
XM-76-1 × Th	91	72	72	63	64	58
TA-622-3	88	73	68	59	64	53
CA-6	92	74	70	65	64	55
TA-700-1 × Ap	89	70	69	59	63	56
TA1001-2	86	69	69	59	63	53
TA-622-2 × Ap	89	68	70	63	67	56
TA-296-3	91	67	67	59	63	51
Regent	82	63	68	58	60	54
Thatcher	83	65	62	54	58	48
Sig. diff. at the 5% level		5.9	4.6	4.5	4.4	5.2

\* Lot I, Lot IIA and Lot IIB—results of 10 days exposure.

\*\* See abbreviations given under Table I.

The agreement between Lot III results and those of Lots I and II suggests a relationship between seed bleaching and earliness of maturity. Correlations on this relationship were worked out for every test in 1944 and 1945 and in all cases they were significant and positive the lowest *r* value being .56.

The results of 2 years of testing using 4 treatments and 16 varieties and lines are given in Table 6. The results on Lot I in 1944 are in close agreement with the 1945 results, and with the Lot IIC results for the 2 years. The Lot IIC results are very consistent, the greatest variation shown by any one variety being only 3%. The Lot IIA results are not as consistent as those for Lot I and Lot IIC. In 1944 Lot IIA encountered more wet spells than the Lot IIA material of 1945, consequently, all scores are lower except those for Apex, MA-163 and Regent. In Lot IIB the



TABLE 5.—THE SIGNIFICANCE OF 14 INTERACTIONS BETWEEN DIFFERENT LOTS AND BETWEEN VARIOUS DATES WITHIN LOTS OF TEST V IN 1945

Interaction	F value	5%	1%
1. Lot I 3 vs. 10 days	0.62	1.55	1.85
2. Lot I 3 vs. 17 days	.95	1.55	1.85
3. Lot I 3 vs. 24 days	1.07	1.55	1.85
4. Lot I 10 vs. 17 days	.44	1.55	1.85
5. Lot I 10 vs. 24 days	.56	1.55	1.85
6. Lot I 17 vs. 24 days	.27	1.55	1.85
7. Lot IIA 10 vs. 20 days	.23	1.55	1.85
8. Lot IIB 10 vs. 20 days	.22	1.55	1.85
9. Lot I (10 days) vs. Lot IIA (10 days)	1.05	1.55	1.85
10. Lot IIA (10 days) vs. Lot IIB (10 days)	.37	1.55	1.85
11. Lot I (10 days) vs. Lot IIC	.85	1.55	1.85
12. Lot IIA (10 days) vs. Lot IIC	.63	1.55	1.85
13. Lot I (10 days) vs. Lot III	.63	1.55	1.85
14. Lot IIA (20 days) vs. Lot IIB (20 days)	.58	1.55	1.85

weather conditions were reversed resulting in lower scores for all varieties in 1945 with the exception of Newthatch. While these variations are not excessively large, they do show the possibility of introducing slight errors in scoring when using the methods outlined for Lots IIA and IIB.

Tables 7 and 8 show a very close agreement between the results of laboratory trials and the field results. In all tests XM-4, Garnet, Marquis and Huron ranked high in resistance, while Thatcher, Regent and TA1001-2 ranked low in resistance to seed bleaching. It is of interest to note that all combinations of wetting and drying produced very similar results with the exception of Tests 4, 11 and 14 given in Table 7. In Test 4

TABLE 6.—COMPARISON OF RESULTS ON 16 WHEAT VARIETIES AND HYBRID LINES FROM 4 TREATMENTS OVER A 2 YEAR PERIOD†

Variety	Lot I, 10 days		Lot IIA, 10 days		Lot IIB, 10 days		Lot IIC, dryer	
	1944	1945	1944	1945	1944	1945	1944	1945
XM-4*	85	82	75	80	77	73	73	74
Marquis	83	81	77	78	77	74	74	76
Huron	83	80	74	81	72	69	73	71
Red Bobs	83	82	76	77	76	71	74	73
XM-76	84	78	73	81	75	69	70	71
MA-261	82	77	72	79	74	67	71	68
MA-131-1	81	76	73	77	75	63	70	67
Henry	80	71	72	73	73	69	72	69
Apex	77	74	71	70	70	60	68	66
MA-163	80	74	69	69	68	63	67	66
TA-622-2 × Ap	76	70	70	73	69	63	66	67
Regent	75	68	65	65	65	59	65	64
Cadet	76	71	68	69	66	61	65	66
TA-1001-2	74	69	67	69	65	58	64	63
Newthatch	72	66	62	65	58	58	58	59
Thatcher	74	67	62	63	61	54	60	58

† The figures in this table were obtained from three 1944 tests and two 1945 tests and for each year were brought to a comparable basis.

\* See abbreviations given under Table I.

the high temperature of 270° F. blistered the samples thus tending to distort their appearance and score. Boiling water, used in Test 14 caused swelling and later shrivelling and cracking of the bran to such an extent that scores could not be taken. In Test 11 the 95% humidity was too high to permit drying before sprouting occurred, therefore, scores could not be taken.

TABLE 7.—THE SUMMARIZED\* RESULTS OF 23 DIFFERENT LABORATORY TESTS ON 10 VARIETIES AND HYBRID LINES OF WHEAT FOR THE RETENTION OF SEED COLOR

Group symbol	Treatment		Variety or line**										
	Detailed procedure		Test No.	XM-4	Garnet	Marquis†	Huron	TA-622-2 X Ap	TA-418-1	TA-357	TA-1001-2	Regent	Thatcher
A	Wet for 10 minutes and dried for 30 minutes at	84° F.	1	79	79	78	68	65	67	60	59	64	59
		175° F.	2	82	81	79	71	69	70	65	66	61	52
		200° F.	3	83	79	82	76	71	74	68	67	59	58
		270° F.	4	78	69	76	73	69	69	57	77	69	56
B	Dried at 200° F. after wetting for	10 min.	5	83	79	80	76	75	74	68	67	59	58
		30 min.	6	81	74	78	74	69	71	67	66	62	60
		75 min.	7	85	79	81	70	64	68	68	67	64	52
C	Wet for 15 minutes then air-dried at the humidity of	25%	8	88	85	82	79	77	77	74	76	73	67
		30%	9	88	85	88	81	77	78	74	73	73	65
		70%	10	89	88	89	80	75	73	74	72	74	71
		95%	11	—	—	—	—	—	—	—	—	—	—
D	Dried at 200° F. after wetting for 10 minutes with water temperatures of	55° F.	12	86	84	87	84	76	77	73	70	69	67
		180° F.	13	81	82	80	80	74	76	71	63	64	58
		212° F.	14	—	—	—	—	—	—	—	—	—	—
E	Wet for 10 minutes and dried at 200° F. Repeated four times.	1	15	82	85	78	75	72	74	72	70	68	55
		2	16	68	70	71	68	66	64	58	58	60	52
		3	17	66	64	63	64	60	58	55	55	55	50
		4	18	58	56	59	55	51	55	52	51	50	47
	Field results (10 days exposure), also Lot III and IIC results.‡	Lot I	19	82	—	81	80	72	75	73	68	63	64
		Lot IIA	20	77	—	77	78	70	75	72	69	68	61
		Lot IIB	21	70	—	74	68	63	64	64	58	58	54
		Lot III	22	66	—	71	65	57	59	55	53	54	48
		Lot IIC	23	73	—	72	71	67	65	64	63	60	58

\* Each result is the average of duplicate samples.

\*\* See list of abbreviations given under Table I.

† The field results for Marquis were obtained by interpolation from the 1945 results of the Coop. Test of New Wheats.

‡ The variety Garnet was not included in the field tests.

The close agreement between the results of the different tests makes the selection of a desirable laboratory technique mostly a matter of convenience. Where a dryer, which can be operated at controlled temperatures, is available, Test 3 is most suitable, since a large number of samples can be tested in a very short time. If a dryer is not available Tests 7, 8 and 9 are quite suitable, although the drying time of the samples will range from 12 to 20 hours respectively, for each test.



TABLE 8.—THE RANKING OF 10 WHEAT VARIETIES AND LINES ACCORDING TO THEIR RESISTANCE TO SEED BLEACHING AFTER BEING SUBJECTED TO 23 DIFFERENT TESTS.

Group symbol	Treatment		Variety or lines*										
	Detailed procedure		Test No.	XM-4	Garnet	Marquis	Huron	TA-622-2 X Ap	TA-418-1	TA-357	TA-1001-2	Regent	Thatcher
A	Wet for 10 minutes and dried for 30 minutes at	84° F.	1	1	1	3	4	6	5	8	9	7	10
		175° F.	2	1	2	3	4	6	5	8	7	9	10
		200° F.	3	1	3	2	4	6	5	7	8	9	10
		270° F.	4	1	6	3	4	6	6	9	2	6	10
B	Dried at 200° F. after wetting for	10 min.	5	1	3	2	4	5	6	7	8	9	10
		30 min.	6	1	3	2	3	6	5	7	8	9	10
		75 min.	7	1	3	2	4	8	5	5	6	8	10
C	Wet for 15 minutes then air dried at the humidity of	25%	8	1	2	3	4	5	5	8	7	9	10
		30%	9	1	3	1	4	6	5	7	9	9	10
		70%	10	1	3	1	4	5	7	6	9	8	10
		95%	11	—	—	—	—	—	—	—	—	—	—
D	Dried at 200° F. after wetting for 10 minutes with water at temperatures of	55° F.	12	2	3	1	3	6	5	7	8	9	10
		180° F.	13	2	1	3	3	6	5	7	9	8	10
		212° F.	14	—	—	—	—	—	—	—	—	—	—
E	Wet for 10 minutes and dried at 200° F. Repeated four times	1	15	2	1	3	4	6	5	6	8	9	10
		2	16	3	2	1	3	5	6	7	7	9	10
		3	17	1	2	4	2	5	6	8	8	8	10
		4	18	2	3	1	4	7	4	6	7	9	10
	Field results (10 days exposure), also Lot IIC and Lot III results	Lot I	19	1	—	2	3	6	4	5	7	8	9
		Lot IIA	20	1	—	2	3	6	4	5	7	8	9
		Lot IIB	21	2	—	1	3	6	4	4	8	8	9
		Lot III	22	2	—	1	3	5	4	6	8	7	9
		Lot IIC	23	1	—	2	3	4	5	6	7	8	9

\* See list of abbreviations given under Table 1.

\*\* The variety Garnet was not included in the field tests.

## DISCUSSION

This study has shown that resistance to seed bleaching is a definite varietal characteristic with wide and stable differences between varieties. The hybrid lines of Marquis crossed with Apex or Apex sibs reacted within the parental range but several of the lines from the cross Thatcher × Apex excelled Apex, the more resistant parent. The results indicate that the expectation for seed bleaching resistance is about the same as for most quantitative characteristic, viz., within the parental range with possibilities of transgressive segregation.

In the four tests conducted in 1944 and the two tests conducted in 1945, resistance to seed bleaching was found to be correlated with lateness of maturity. This largely accounts for the close agreement between the Lot I and Lot III results given in Table 4, since the resistant late varieties benefited from fewer days of exposure while the early varieties, although lower in resistance, were exposed to the weather for a longer time.

Since there is close agreement between the results of the bleaching tests carried on in the field under natural weather conditions and those carried on in the laboratory, either type of test may be used as a practical method of determining varietal resistance to seed bleaching. Owing to the labour and expense involved in field tests the laboratory tests for bleaching resistance would seem to be preferable.

### SUMMARY

A total of 152 varieties and purified hybrid lines were tested for their resistance to seed bleaching under both natural and laboratory conditions at Saskatoon in 1944 and 1945. Under natural weather conditions the varieties were tested both as unthreshed heads and as threshed seeds.

Significant differences in resistance to seed bleaching were found between the varieties tested.

Close agreement was found between the results produced by natural weathering on unthreshed and threshed samples and those results obtained by the use of a laboratory technique.

Wetting and drying was the principle involved in the laboratory tests. Wetting the samples for ten minutes and then drying them either in a dryer at 200° F. or in the open air at room temperature seemed to be the most satisfactory.

Resistance to seed bleaching was found to be related to lateness of maturity.

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# SCLEROTINIA SATIVA, AND RELATED SPECIES, AS ROOT PARASITES OF ALFALFA AND SWEET CLOVER IN ALBERTA<sup>1</sup>

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Several fungi have been found associated with root rot of alfalfa and sweet clover in Alberta. A detailed study was previously made of *Plenodomus Meliloti*, by Sanford (13), and of the related groups *Cylindrocarpon* spp. (5), and *Fusarium* spp. (6). Attention was also paid to *Sclerotinia sativa* Drayton and Groves (9) in studies on root invasion (4), and varietal resistance (7). The present paper deals mainly with continued studies on the latter species, especially with regard to prevalence, pathogenicity, host range, persistence, and cultural characteristics. Related species included for purposes of comparison are, *S. sclerotiorum* (Lib.) De Bary, a *Botrytis* of the *cinerea* type (recently renamed as *Botryotinia Fuckeliana* by Whetzel (14)), *S. Trifoliorum* Erikss., and *S. minor* Jagger. The two latter fungi, although not yet found here, cause damage to legume forage crops in other regions.

## OCCURRENCE IN ALBERTA

The results of survey and isolation studies made in Alberta during the past 15 years indicate that *Sclerotinia* root rot of sweet clover and other legume forage crops is most frequently caused by the species now known as *S. sativa*. The damage was usually ascribed to *S. Trifoliorum* (12) until 1934, when the late Dr. H. H. Whetzel kindly examined several isolates from Alberta and expressed the opinion that they were distinct from previously described species. At that time the fungus was referred to as *Sclerotinia* sp. (4). However, Drayton and Groves (9), who succeeded in obtaining mature apothecia from cultures isolated from legumes and tulips, described it as a new species, *S. sativa*, in 1943 (9).

Although sometimes very destructive to sweet clover in Alberta, *S. sativa* does not occur very commonly and is probably not as important as *Cylindrocarpon Ehrenbergi* and other root-rotting pathogens previously studied (7). It is found in sweet clover fields in various sections of the province nearly every year, causing damage which varies from a trace to 50% of the plants. It has also been isolated from diseased sweet clover specimens sent from Saskatchewan. There is no indication that the disease is spreading or increasing in importance in any of the localities in which it has been found. The damage is usually confined to individual fields and does not increase unless sweet clover is re-planted or allowed to volunteer in infested fields. Occasionally the pathogen has been isolated from slightly damaged roots of alfalfa in Alberta. In other regions it has been reported as the cause of a destructive bulb rot of tulip and narcissus (9).

Under natural conditions the root rot caused by *S. sativa* has been found only in the early spring. Affected sweet clover plants usually fail to grow, or they produce very weak shoots. The roots may be rotted to varying degrees and are often completely decayed (Plate 1, A, B, and C).

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The tissues of severely rotted roots are at first soft and watery and usually covered with the cottony white mycelium of the fungus. As the soil warms up this mycelium is replaced by black sclerotial bodies of varying size, and the decayed tissues become dry and shredded, and soon disintegrate (Plate 1, D). In alfalfa stands root-rot damage caused by *S. sativa* is seldom severe enough to affect the growth of the plants. The light brown, superficial lesions produced on the upper part of the root seldom contain sclerotia (Plate 1, E), and can be diagnosed only by means of isolation.

*S. sclerotiorum* also causes a root rot of alfalfa and sweet clover in Alberta, but, unlike *S. sativa*, it is a warm weather parasite causing damage only during the growing season. Sweet clover is more commonly affected than alfalfa, but there is seldom serious damage to either crop. The plants are killed singly or in small patches, and the symptoms on the roots are very similar to those produced by *S. sativa*, except that the sclerotia on them are generally larger and develop more rapidly. Sclerotia of *S. sclerotiorum* have been found in several samples of alfalfa seed grown in Alberta and Saskatchewan, but they were not observed in the stems of the diseased plants. Bisby (1), however, reported finding occasional sclerotia of this species in the stems of legumes in Manitoba. Another possible source of the sclerotia in these seed samples is from the stems of Canada Thistle or other susceptible weeds growing in the alfalfa and threshed with it. During the present study *S. sclerotiorum* was isolated from diseased sunflowers, beans, carrots, parsnips, lettuce, and tomatoes. On these hosts it caused more damage than on alfalfa or sweet clover.

Neither *S. Trifoliorum* nor *S. minor* have been found on legume forage crops or other hosts in Alberta. Moreover, stem rot commonly produced on legumes in other regions (10) has never been observed here.

*Botrytis* sp. (*cinera* type) is frequently isolated from diseased roots of alfalfa and sweet clover in Alberta, where it usually occurs in lesions with *Cylindrocarpon Ehrenbergi* (5) or other root-rotting pathogens. Since preliminary tests indicated that this *Botrytis* could parasitize the roots of alfalfa and sweet clover under certain conditions, several isolates from different sources were included in the present investigation.

## INFECTION STUDIES

### PATHOGENICITY TESTS ON ALFALFA AND SWEET CLOVER

Isolates of *Sclerotinia sativa*, *S. sclerotiorum*, *S. minor*, *S. Trifoliorum*, and *Botrytis* sp. from various hosts and sources were tested for pathogenicity on the roots of Grimm alfalfa and Arctic sweet clover. All tests were made in the field on growing plants during the summer, as well as on dormant plants in the winter. Certain isolates of each species, indicated in Table 1, were included in 5 winter tests. Plants about 1 year old were inoculated by placing oat-hull inoculum in contact with the roots in a shallow trench dug along the side of the row. For the winter tests, the plants were inoculated in the late fall and dug about the time that growth started the following spring. Notes on the summer tests were taken about 4 weeks after the plants were inoculated. The infection rating given each plant was expressed in percentage (5).



As indicated by the results of the winter tests summarized in Table 1, *S. sativa* severely attacked the roots of the dormant plants. All isolates tested were equally destructive to sweet clover, and those from tulip bulbs produced severe rotting of alfalfa. However, during summer there was only slight infection on growing plants of sweet clover, and alfalfa usually escaped. The symptoms produced in the early spring were similar to those previously described, except that the isolates from tulip bulbs caused extensive lesioning of alfalfa roots, as compared to the slight damage caused by the isolates from legumes (Plate 1, E).

TABLE 1.—RELATIVE VIRULENCE OF ISOLATES OF DIFFERENT SPECIES OF *Sclerotinia* AND *Botrytis* ON ROOTS OF ALFALFA AND SWEET CLOVER

Species	Isolate		Infection Rating <sup>1</sup> , %					
	No.	Source	Alfalfa			Sweet clover		
			Summer	Winter	Winter <sup>2</sup>	Summer	Winter	Winter <sup>2</sup>
<i>Sclerotinia sativa</i>	1	Alfalfa	6	33		26	100	
	2	White sweet clover	7	25	23	22	100	96
	3	White sweet clover	6	66		23	100	
	4	White sweet clover	7	39		34	100	
	5	Yellow sweet clover	7	36		31	100	
	6	Tulip bulbs (New York)	6	92	95	21	100	100
	7	Tulip bulbs (Quebec)	5	89		17	100	
	8	Tulip bulbs (Quebec)	7	58		33	98	
<i>S. sclerotiorum</i>	1	Alfalfa	11	11		86	35	
	2	Yellow sweet clover	20	12	26	100	53	56
	3	Carrot	30	24		84	92	
	4	Lettuce	17	22		70	40	
	5	Snap bean	9	30	34	93	64	71
	6	Tomato	7	30		96	60	
<i>S. minor</i>	1	Lettuce (New York)	13	25	21	91	39	36
	2	Chicory (Belgium)	70	30	39	100	87	93
	3	Potato (Quebec)	5	5		6	8	
	4	Sunflower (Quebec)	6	5		6	5	
<i>S. Trifoliorum</i>	1	Red Clover (Kentucky)	41	100	92	95	100	100
	2	Clover (England)	70	39		92	100	
	3	Broad bean (England)	54	61		96	60	
	4	Crimson clover (Germany)	100	96		100	100	
	5	Sweet clover (British Columbia)	94	82		100	100	
<i>Botrytis</i> sp. ( <i>cinerea</i> type)	1	Alfalfa	20	4		25	20	
	2	Sweet clover	6	7	10	15	25	29
	3	Sweet pea	28	9		15	15	
	4	Broad bean	4	24		5	32	
	5	Potato	8	10		12	10	
	6	Wheat seed	9	13		8	32	
Control			6	5	3	9	6	4

<sup>1</sup> Average rating of 20 plants.

<sup>2</sup> Five-year average.

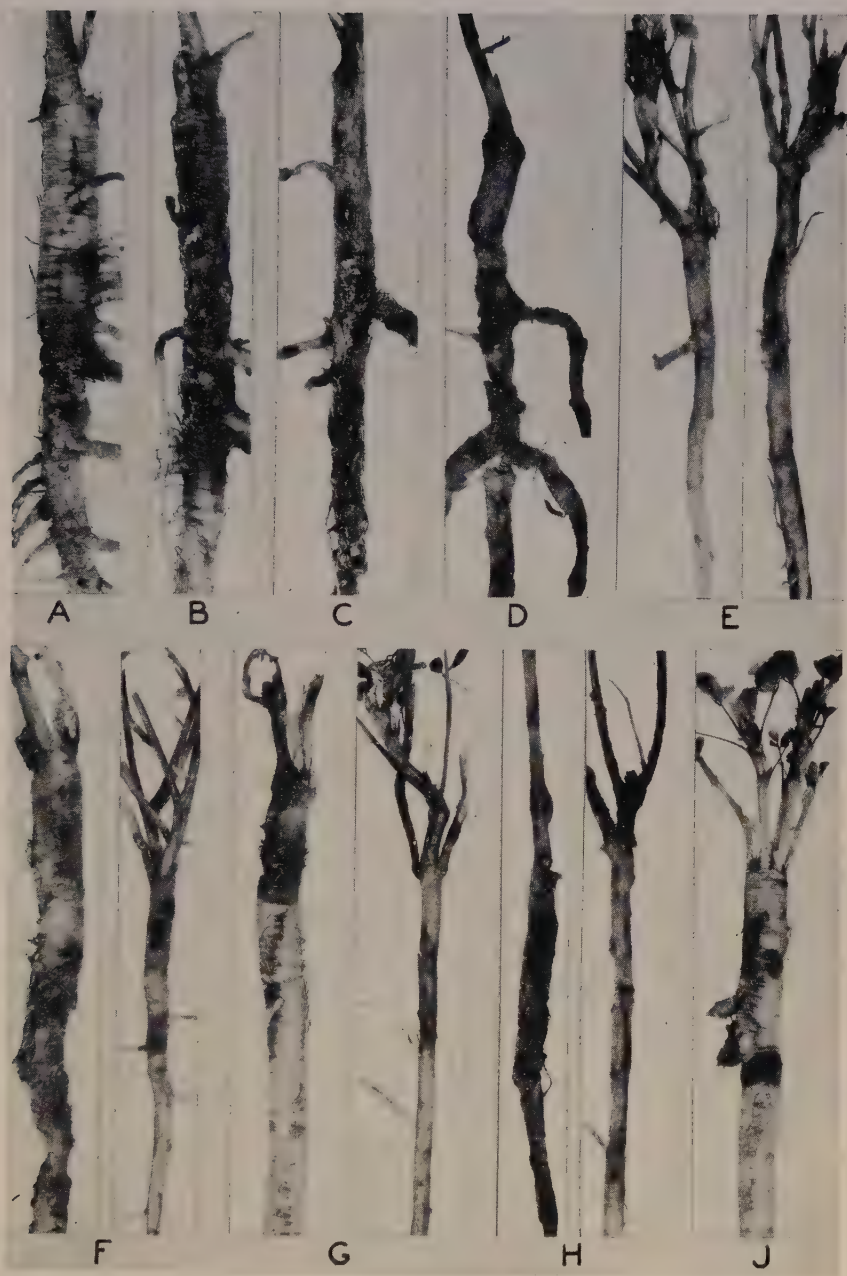


PLATE 1, A to D. Progressive symptoms produced by *Sclerotinia sativa* on roots of sweet clover in the early spring. E. Alfalfa roots attacked by *S. sativa* (left) from sweet clover, and (right) from tulips. F, G and H. Sweet clover (left), and alfalfa (right) attacked by; (F) *S. sclerotiorum*; (G) *S. minor*; and (H) *S. trifoliorum*. J. Sweet clover attacked by *Botrytis* sp. (*cinerea* type).



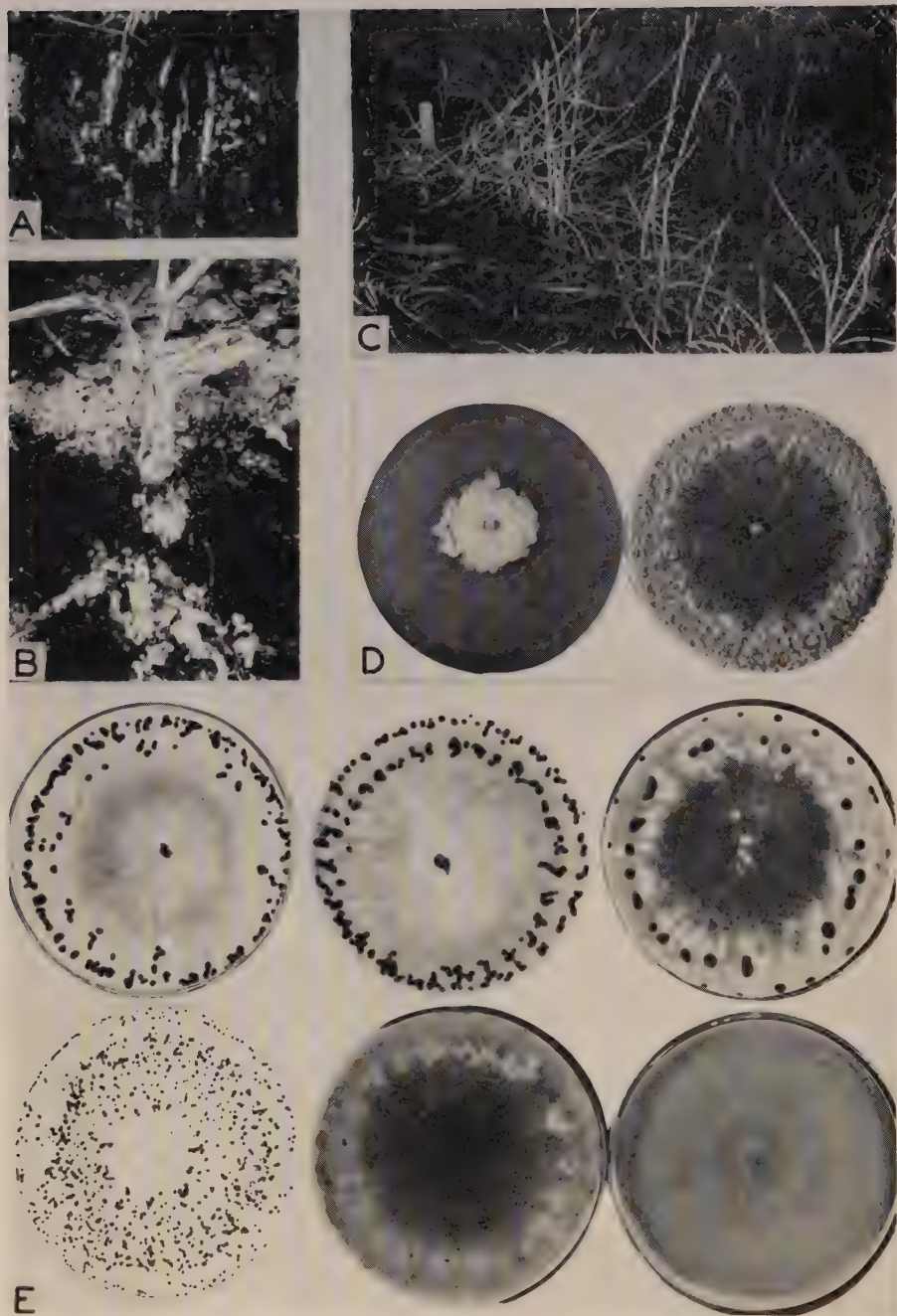


PLATE 2, A and B. Mycelium of *Sclerotinia sativa* on roots of sweet clover, attacked in partially frozen soil during early spring. C. Killing by *S. sativa* of sweet clover seeded in infested land after 8 years of continuous fallow. D. Growth of *S. sativa* on media made from roots of alfalfa (left), and sweet clover (right). E. Week-old colonies of species of *Sclerotinia* and *Botrytis* on potato dextrose agar (left to right). Above: *S. sativa* isolated from sweet clover, and from tulip; *S. sclerotiorum*. Below: *S. minor*, *S. Trifoliorum*, *Botrytis* sp. (*cinerea* type).

*S. sclerotiorum* was generally more virulent during the summer than in the winter tests (Table 1). In fact, periodic examinations indicated that most of the damage caused by this species in the winter tests actually occurred after the soil started to warm up prior to the time of note-taking in early May, rather than at the time of the first spring thaw, as in the case of *S. sativa*. Isolates of *S. sclerotiorum* from alfalfa, sweet clover, carrot, lettuce, snap bean, and tomato proved pathogenic in varying degrees, but all caused much more damage to sweet clover than to alfalfa (Plate 1; F).

The cultures of *S. minor* from lettuce and chicory proved highly virulent to sweet clover, especially in the summer tests, and caused slight to moderate damage to alfalfa (Table 1). The other two cultures tested were non-pathogenic on both hosts.

All cultures of *S. Trifoliorum* tested were extremely virulent and usually caused complete rotting of sweet clover roots and moderate to severe damage to those of alfalfa in both summer and winter tests (Table 1). Stem rot symptoms were not produced by this species or by any of the others studied.

The symptoms of root attack by *S. sclerotiorum*, *S. minor*, and *S. Trifoliorum* (Plate 1, F, G, and H) were similar to those described for *S. sativa*, except for the variations in severity indicated. The sclerotia produced by all 4 species on decayed roots of sweet clover varied greatly in number and size and were seldom of diagnostic value. Severe rotting of alfalfa roots, accompanied by sclerotial production, was caused only by *S. Trifoliorum* and the tulip isolate of *S. sativa*.

#### CULTIVATED HOSTS

The host range of selected isolates of *Sclerotinia sativa*, *S. sclerotiorum*, *S. minor*, *S. Trifoliorum*, and *Botrytis* sp. was studied by inoculating the following forage and vegetable crops and varieties: alfalfa, *Medicago sativa* L. (Grimm), and *M. falcata* L.; sweet clover, *Melilotus alba* Desr. (Arctic), and *M. officinalis* (L.) Lam. (Common Yellow); red clover, *Trifolium pratense* L. (Altaswede); alsike clover, *T. hydridum* L.; sunflower, *Helianthus annuus* L. (Mennonite); broad bean, *Vicia Faba* L. (Windsor); bush bean, *Phaseolus vulgaris* L. (Tendergreen); lettuce, *Lactuca sativa* L. (Cosberg); tomato, *Lycopersicon esculentum* Mill. (Bounty); carrot, *Daucus Carota* L. (Chantenay); parsnip, *Pastinaca sativa* L. (Hollow Crown); cabbage, *Brassica oleracea* L. var. *capitata* L.; turnip, *Brassica Rapa* L.

Field tests were made during the summer and winter as previously described. In addition, the ability of the different isolates to cause storage rot of carrot, parsnip, cabbage, and turnip was studied. Sound specimens were inoculated by placing a small portion of mycelial inoculum in a slight wound. Duplicate tests were made at 5° and 15° C. for periods of 3 weeks and 10 days, respectively. At least 10 inoculations were made with each isolate on all hosts in each test.

*S. sativa* consistently caused more damage in winter than during the summer (Table 2). As in previous studies (7), *Medicago falcata* was more resistant than common alfalfa (*M. sativa*), and Common Yellow sweet



TABLE 2.—RELATIVE SUSCEPTIBILITY<sup>1</sup> OF CERTAIN PLANTS TO ATTACK BY SPECIES OF *Sclerotinia* AND *Botrytis*

Host plant	Tested	<i>S. sativa</i>		<i>S. sclerotiorum</i>	<i>S. minor</i>	<i>S. Tri- foliorum</i>	<i>Botrytis</i> sp.	Control
		Sweet clover <sup>2</sup>	Tulip <sup>2</sup> bulbs	Sweet clover <sup>2</sup>	Lettuce <sup>2</sup>	Red clover <sup>2</sup>	Sweet clover <sup>2</sup>	
Alfalfa— <i>M. sativa</i>	Summer	T	T	L-M	L	M	T	T
	Winter	L-M	M-S	L	L-M	S	L	T
Alfalfa— <i>M. falcata</i>	Summer	O	T	L	L	M	T	T
	Winter	T-L	M	L	L	S	T	T
Sweet clover— <i>M. alba</i>	Summer	L	L	S	S	S	L	T
	Winter	S	S	M	M	S	L-M	T
Sweet clover— <i>M. officinalis</i>	Summer	L	L	S	S	S	L	T
	Winter	M	M-S	M	M	S	L-M	T
Red clover	Summer	T	T	M	L-M	S	L	T
	Winter	L-M	L-M	L	L-M	S	L	T
Alsike clover	Summer	T	T	L-M	L-M	S	L	T
	Winter	L	L	L	L-M	S	L	T
Sunflower	Summer	T	L	S	S	M	T	O
Broad bean	Summer	T-L	T-L	L	L	M	L	O
Bush bean	Summer	L	L	L	L	L-M	T	T
Lettuce	Summer	L	L	S	M	L	T	O
Tomato	Summer	O	O	L	L-M	L	T	O
Carrot	Summer	T	T	M	L	T	O	O
	Storage	T-L	T-L	S	L	T	T	T
Parsnip	Summer	O	O	M	L	T	O	O
	Storage	T	T	S	L	T	T	O
Cabbage	Storage	T	T	S	T	O	O	O
Turnip	Storage	T	T	S	L	O	O	T

<sup>1</sup> O—none; T—trace; L—light; M—moderate; S—severe.<sup>2</sup> Source of isolate.

clover (*Melilotus officinalis*) was less severely infected than Arctic (*M. alba*). The roots of red clover and alfalfa were about equally susceptible, but those of alsike clover were only slightly damaged. In summer tests *S. sativa* was weakly parasitic on sunflower, beans, and lettuce, and did not attack tomato or parsnip. Moderate rotting, however, occurred in inoculated parsnips overwintered in the field. There was only a trace to light infection on parsnips, carrots, cabbages, and turnips stored at 5° and 15° C. The isolates from sweet clover and tulip bulbs caused a similar degree of infection on all hosts except alfalfa. As reported by Drayton and Groves (9), isolates from both sources caused severe rotting of tulip bulbs in winter tests.

*S. sclerotiorum* proved pathogenic to varying degrees on all hosts studied (Table 2). Root infection was moderate to severe in sweet clover, and slight to moderate in alfalfa, red clover, and alsike clover. All of these crops were more severely attacked by this species during the summer

than in the winter tests. Infection was severe on sunflower and lettuce, but only light on broad bean, snap bean, and tomato. Carrot, parsnip, cabbage, and turnip, stored at 5° and 15° C. were severely rotted, but infection was only moderate on growing roots of carrot and parsnip in the field. *S. sclerotiorum* has been reported as parasitic on a wide range of host plants in the temperate regions (1, 2, and 15).

*S. minor* caused about the same degree of infection as *S. sativa* on the legume forage crops in the winter tests, but usually it was more virulent during the summer (Table 2). In the field tests, infection was severe on sunflower, moderate on lettuce, light to moderate on tomato, and light on broad bean, bush bean, carrot, and parsnip. Little or no damage was caused to carrot, parsnip, cabbage, and turnip stored at 5° and 15° C. *S. minor* has been previously reported on clover (3) and on various vegetables (11).

*S. Trifoliorum* was more virulent on legumes than any of the other species tested (Table 2). In all tests it caused severe rotting of the roots and death of sweet clover, red clover, and alsike clover. In the case of alfalfa, both *Medicago sativa* and *M. falcata* were killed in the winter tests, and moderately damaged during the summer. Infection was moderate in broad beans, and light to moderate in bush beans. The damage was moderate in sunflower but was absent or slight in the other non-leguminous plants studied in the field and in storage. *S. Trifoliorum* is best known as the cause of stem rot of clovers (10), and is seldom reported on other hosts.

*Botrytis* sp. caused slight damage to forage crop legumes and broad bean. On the other hosts studied there was only a trace of infection or none, as in the controls. This fungus has been previously reported on alfalfa and clovers (3), chiefly as the cause of grey mould of the shoots and blossoms. It occurs commonly as a weak parasite or saprophyte on the dead or dying parts of many other plants.

#### WILD HOSTS

The host range of *S. sativa* among native plants and perennial weeds was determined by placing inoculum against the roots *in situ* in the late fall. The inoculated plants and controls were dug up for examination the following spring. The relative susceptibility of the plants studied, based on the average results obtained in 2 successive winter tests with each, was as follows:—Highly susceptible; *Achillea Millefolium* L., *Artemisia gnaphalodes* Nutt., *Aster conspicuus* Lindl., *Cirsium arvense* (L.) Scop., *Cirsium undulatum* (Nutt.) Spreng., *Helianthus* sp., *Lactuca pulchella* (Pursh) D C., *Plantago major* L., *Senecio eremophilus* Richards, *Solidago Canadensis* L., *Sonchus arvensis* L., *Taraxacum officinale* Weber, and *Urtica procera* Muhl.; moderately susceptible: *Aster laevis* L., *Heracleum lanatum* Michx., and *Potentilla* sp. Slightly susceptible; *Agastache anethiodora* (Nutt.) Britt., *Argentina Anserina* (L.) Rydb., *Chamaenerion spicatum* (Lam.) S. F. Gray, and *Vicia americana* Muhl.

Owing to decay in the controls, inconclusive results were obtained with several other native plants, but all appeared to be susceptible in some degree to attack by *S. sativa*. This pathogen has not yet been isolated from naturally infected roots of any of these plants, although its potential host range is wide. All of the perennial weeds inoculated, including Canada



thistle (*Cirsium arvense*), sow thistle (*Sonchus arvensis*), and dandelion (*Taraxacum officinale*), were severely damaged, but control by this means does not seem practical. These weeds have been previously reported as hosts of *S. sclerotiorum* (15).

No infection was obtained in winter tests of *S. sativa* on winter wheat, winter rye, and several cultivated and native species of the grass genera *Agropyron*, *Bromus*, *Festuca*, *Phleum*, and *Poa*.

### FIELD STUDIES ON *S. SATIVA*

#### DISEASE DEVELOPMENT

The progress of infection of sweet clover roots by *S. sativa* in the early spring was studied in the field by periodic examination of plants grown in infested soil. These plants grew normally during the first season, and there was no evidence of infection until the following spring. Observations were made at the time of the first thaw by baring a few roots on one side to a depth of about 4 inches. Thereafter examinations were made daily while the disease was progressing rapidly. Another method that gave good results was to make these observations on roots of inoculated plants in boxes of soil. These were brought in and the soil thawed out slowly at a temperature near freezing in a refrigerated room.

The first signs of infection appeared on the roots in the form of small watery areas shortly after thawing started. These decayed areas developed very rapidly and often involved the entire root system within less than a week. By this time the decayed roots were usually covered with the cottony white mycelium of the fungus (Plate 2, A and B). This mycelium followed the rootlets from plant to plant and penetrated to a very limited extent into the surrounding soil. Even at this stage the soil was often still partially frozen. When it warmed up and growth started in the control plants, the mycelium was replaced by sclerotia and the decayed tissues became dry and shredded.

#### SPREAD OF PATHOGEN IN SOIL

Since the early spring observations indicated that *S. sativa* spreads mainly from one plant to another by means of contiguous roots or rootlets, a field study was made to determine if the fungus could grow directly through the soil. In the late fall, inoculum was placed in trenches about 2 inches deep, at varying distances from rows of sweet clover. These trenches were dug parallel to the rows and all roots and rootlets were removed from the intervening soil. In the following spring, infection occurred on all plants having roots in actual contact with inoculum. When the inoculum was  $\frac{1}{4}$  inch distant from the roots, the average infection was 42%, and when 1 inch, 5%. At greater distances no damage occurred. However, when inoculum was placed in a closely planted row or stand the pathogen spread and killed the plants to a distance of at least 6 inches. Similar results were obtained with inoculum placed on the surface of the soil, except that usually the fungus spread to a lesser extent above than below ground. Davis (8) found that *S. sclerotiorum* progressed only 5 cm. from the centre of infection on the surface of the soil and that healthy cabbage plants were infected by contact with diseased individuals.

## PERSISTENCE IN SOIL

Field studies were made on the ability of *S. sativa* to persist in the soil in the absence of a host plant. A plot at Edmonton, in which sweet clover was severely damaged by *S. sativa* in the early spring of 1937, was subsequently fallowed, except for successive portions replanted each year to sweet clover. These plantings all grew well during the first season, but those of the first 4 years were completely killed out by *S. sativa* in the early spring of the second season. Since then a small portion of the plants, increasing from 5% in 1942 to 15% in 1945, have survived each spring. These plants were weakened, however, by partial rotting of the roots, and the fungus still caused extremely severe damage in the early spring of 1945 in the portion of the plot that had been summer-fallowed for 8 years (Plate 2, C). These results have been confirmed in other plots and fields where it has been found impossible to maintain a new stand of sweet clover following root-rot damage caused by *S. sativa*. Annual crops and grasses were unaffected, even when planted immediately in such land, and alfalfa was seldom seriously damaged.

## CULTURAL STUDIES ON SCLEROTINIA SPP.

### LONGEVITY OF SCLEROTIA

Field observations indicated that the sclerotia were probably not responsible for the long continued persistence of *S. sativa* in the soil. Sclerotia were abundant in the diseased root tissues of sweet clover following infection in the early spring, but disappeared as the tissues disintegrated. By September only a few soft fragments of sclerotia remained in the shells of the old decayed roots.

In laboratory experiments, sclerotia of *S. sativa* and *S. sclerotiorum*, in lots of 50, were made up in cheesecloth-covered packets with an equal volume of air-dried soil. These packets were stored at room temperature for 6 months\* in cans of soil at three moisture contents, namely, air dry, about 55%, and 70 to 80% m.h.c., respectively. Within a month the sclerotia of *S. sativa* stored in the moist and wet soils were almost completely decomposed and unidentifiable. Those stored in the dry soil remained sound for the duration of the experiment. The sclerotia of *S. sclerotiorum* were also decomposed in a month in the wet soil, but did not start to decay until after 6 months in the moist soil. In dried herbarium specimens sclerotia of both species retained their original form indefinitely. Sclerotia of *S. sativa* failed to produce mycelium after 3 years storage, while those of *S. sclerotiorum* still remained viable after 7 years. Brown and Butler (2) noted the general tendency of sclerotia of *S. sclerotiorum* to decay in moist soil, but found that they could live for 11 years under dry conditions.

Apothecia and ascospores of *Sclerotinia* spp. have not been observed under natural field conditions in Alberta, and apparently do not aid in the dispersal of these fungi as is the case in warmer, more humid regions (2, 10). Apothecia of *S. sativa* have not been reported in nature. However, they were obtained by Drayton and Groves (9) from cultures in the laboratory and greenhouse.



### INFLUENCE OF MEDIA ON GROWTH

To check on host relationships, isolates of *S. sativa*, *S. sclerotiorum*, *S. minor*, and *S. Trifoliorum* were cultured on media made from the extracted root juices of several varieties of alfalfa and sweet clover. These media consisted of 5% root decoction in water agar. Similar results were obtained from the root juices sterilized by steam or passed through a Berkefeld filter. The media from alfalfa roots proved to be much less favourable than those from the sweet clover both for growth and sclerotial formation, which results agree with the data obtained in the infection studies. These differences were particularly marked in the case of the legume isolates of *S. sativa* (Plate 2, D), and occurred to about the same degree when different varieties of alfalfa and sweet clover were used.

Potato-dextrose agar was the best general medium for *S. sativa* and the other species. Fair results were given by Czapek's peptone-dextrose and malt agars, but Dox's inorganic salt, Molisch's salt-peptone, bean-pod, and corn-meal agars were unsatisfactory. In confirmation of field results, little or no growth occurred in natural soil, even when it was steam sterilized. A soil medium, sterilized or non-sterilized, containing about 10% by volume of ground oat hulls or other organic matter, however, favoured rapid growth and was sometimes used for the preparation of inoculum.

A comparative study was made of the cultural characteristics of representative isolates of *Sclerotinia* spp. and *Botrytis* sp. when grown on potato-dextrose agar. As shown in Plate 2, E, the isolates of *S. sativa* from sweet clover and tulips were similar in appearance and produced a whitish, close-growing mycelium and fairly numerous mid-sized sclerotia. In test-tube culture these isolates sometimes developed a fluffy, greyish, aerial mycelium resembling that of *Botrytis* sp. *S. sclerotiorum* and *S. Trifoliorum* produced more aerial mycelium and larger sclerotia than *S. sativa* (Plate 2, E). In *S. Trifoliorum* the sclerotia developed more slowly than in *S. sclerotiorum*, and were often partially covered by the mycelium. Most isolates of *S. minor* produced a scant mycelium and extremely numerous, small sclerotia. All isolates of *Botrytis* sp. developed an abundant, greyish aerial mycelium and no sclerotia, except occasionally when freshly isolated. After being cultured for a long period some isolates, particularly of *S. sativa*, tended to run-out and ceased to produce typical mycelium and sclerotia.

### INFLUENCE OF TEMPERATURE ON GROWTH

Representative isolates of *S. sativa*, *S. sclerotiorum*, *S. minor*, *S. Trifoliorum*, and *Botrytis* sp. were grown in quadruplicate plates of potato-dextrose agar at temperatures ranging from  $-4^{\circ}$  to  $34^{\circ}$  C. The average results obtained are summarized in Table 3.

In the low-temperature series all species grew slowly at  $1^{\circ}$  C., but only *S. sativa* grew at temperatures below freezing. This species grew slowly even at  $-4^{\circ}$  C. on frozen agar, producing a compact colony about 20 mm. in diameter in one month. At temperatures from  $5^{\circ}$  to  $10^{\circ}$  C. sclerotia were produced slowly and sparingly by all species.

*S. sativa* and *S. Trifoliorum* produced most growth at  $17^{\circ}$  C., and apparently both have an optimum below  $20^{\circ}$  C. Best growth of *S. minor* and *Botrytis* sp. occurred at  $20^{\circ}$  C., and of *S. sclerotiorum* at about  $25^{\circ}$  C.

TABLE 3.—INFLUENCE OF TEMPERATURE ON GROWTH OF *Sclerotinia* spp.  
AND *Botrytis* ON POTATO-DEXTROSE AGAR

Species	Average diameter of colony in millimetres									
	10 days					3 days				
	1° C.	5° C.	10° C.	14° C.	17° C.	20° C.	25° C.	28° C.	31° C.	34° C.
<i>S. sativa</i>	7	28	90	25	32	30	25	6	0	0
<i>S. sclerotiorum</i>	5	20	85	59	83	86	90	48	6	0
<i>S. minor</i>	6	18	77	21	25	34	27	7	0	0
<i>S. Trifoliorum</i>	15	19	50	27	29	26	15	0	0	0
<i>Botrytis</i> sp. ( <i>cinerea</i> type)	11	33	77	42	49	53	45	8	0	0

Growth of all the species dropped off rapidly at the higher temperatures, and none of them grew at 34° C. The colonies of *S. Trifoliorum* ceased to develop at 28° C., those of *S. sativa*, *S. minor*, and *Botrytis* sp. at 29° to 31° C., and *S. sclerotiorum* at about 32° C. Growth of *S. sativa*, *S. minor*, and *S. Trifoliorum* was relatively slow at temperatures above 10° C., as compared to that of *S. sclerotiorum*.

#### DISCUSSION

Although the species of *Sclerotinia* studied herein are potentially capable of causing severe root-rot damage in sweet clover, they are not yet very destructive in Alberta. In fact *S. sativa* and *S. sclerotiorum*, the only species found in this region, occur less frequently and cause less general damage than most of the other root- and crown-rotting pathogens previously studied (7). Apparently our climatic conditions do not favour the development and spread of these fungi, perhaps being too cool and dry for the development of the stem-rot symptoms and apothecia commonly produced by *Sclerotinia* spp. in warmer, more humid regions (2, 10). When the above-ground parts of the plants are not infected there is less opportunity for the spread of the mycelium and sclerotia. Extensive distribution of these pathogens is even more effectively prevented when there are no ascospores to be widely disseminated by the wind. Thus, under Alberta conditions, it appears that *Sclerotinia* spp., once established in a given soil, are mainly limited to local spread through contact between the roots of diseased and healthy plants, or by the transfer of infective material during cultural operations. Bisby (1) concluded that climatic conditions in Manitoba did not favour maximum development of the disease caused by *S. sclerotiorum*.

The recently described species *S. sativa* was of particular interest in this investigation. To date it has been found only on roots of alfalfa and sweet clover in Western Canada and on bulbs of tulip and narcissus in Eastern Canada and the United States (9). This limited natural host range will probably be extended in the future, since the present study has shown that a number of other species are susceptible to attack by *S. sativa* during early spring. As a parasite of the dormant plants, it causes damage that may be overlooked or confused with true winter-killing. It would be interesting to know if *S. sativa* is native or introduced. Although it has

not yet been isolated from naturally infected native hosts, apparently it is harboured by a wide range of susceptible plants in nature. When established in a sweet clover field, this species is not active during the growing season, but attacks the dormant plants of sweet clover or other highly susceptible hosts only during the early spring. It also possesses an unusual ability to remain viable in the soil for years, even in the absence of a host. Since its sclerotia are short-lived in moist soil, the fungus apparently persists mainly as a semi-saprophytic mycelium that can live more or less indefinitely on decaying organic matter. Fortunately, many other crops can be safely grown in old sweet clover fields infested with *S. sativa*.

### SUMMARY

*Sclerotinia sativa* is sometimes very destructive to sweet clover in Alberta during the early spring, but it seldom attacks alfalfa and is not as yet very commonly distributed. *S. sclerotiorum* occasionally causes damage to alfalfa and sweet clover during the summer, but it occurs more commonly on sunflowers and vegetable hosts. *S. minor* and *S. Trifoliorum* have not yet been found on any host in Alberta.

*S. sativa* is distinctly a low-temperature parasite of dormant plants. It rapidly invades the roots of its hosts even as the frozen soil thaws during early spring, but its progress is arrested when plant growth begins. In winter inoculation tests in the field it severely attacked sweet clover, but caused slight to moderate damage to alfalfa and red clover, and only slight injury to alsike clover. Parsnip and twenty perennial wild plants were also susceptible. *S. sclerotiorum* was more virulent during summer than early spring and injured sweet clover more than alfalfa, red clover, or alsike clover. *S. minor* attacked the legume forage crops to about the same degree as did *S. sativa*, but it caused most damage during the summer. *S. Trifoliorum* was more virulent on the legume forage crops and beans than any of the other species tested, but did not cause serious damage to any non-leguminous host except sunflower. *Botrytis* sp. (*cinerea* type), frequently associated with root rot of alfalfa and sweet clover in Alberta, was seldom more than weakly virulent on any of the hosts studied.

Excepting *S. sativa*, where the isolates from tulip bulbs were much more virulent to alfalfa than those obtained from legumes, and certain non-pathogenic cultures of *S. minor*, there was little evidence of variation in virulence among the isolates of a given species.

*S. sativa* persisted in fallowed soil and severely damaged sweet clover even after a period of 8 years. However, its sclerotia decayed rapidly in moist soil. No apothecia of this or the other species studied have been found under natural conditions in Alberta.

In pure culture, *S. sativa* produced sclerotia intermediate in size between those of *S. minor* and *S. sclerotiorum*. Alfalfa root media were much less favourable than sweet clover root media, both for growth and sclerotial formation. *S. sativa* and *S. Trifoliorum* grew best at 17° to 19° C., *S. minor* and *Botrytis* sp. at 20° C., and *S. sclerotiorum* at about 25° C. Sclerotial formation was inhibited or retarded in all species at temperatures below 10° C. Growth ceased at about 30° C. in all species.



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# SOME FACTORS AFFECTING APPLE YIELDS IN THE OKANAGAN VALLEY

## IV. ORGANIC MATTER CONTENT OF SOIL<sup>1</sup>

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Both research and practical experience have indicated that organic matter in the soil is essential to successful agriculture. This finding has been applied not only to general crop production, but to tree fruits as well. Investigators in Pennsylvania (1, 5, 9) have found a close relationship between the growth of cover crops and the subsequent performance of fruit trees. This relationship has been attributed primarily to the effects of cover crops in increasing the humus content of the soil. Similar conclusions have been reached by other investigators (4, 6, 10, 11).

The literature on the relationship of humus in the soil to plant and tree performance has been effectively covered by a number of authors (1, 3, 12), and there is no need to cover it again in this paper. Special mention, however, should be made of a report by Cummings (3) in 1937. The organic matter contents of soil samples from 93 Baldwin orchards in New York State were determined by analysing for the total organic carbon and multiplying the results by 1.724. No consistent relationship was found between the organic matter content of the soil and tree yield, or between organic matter and tree size. It was concluded that the organic matter content at any one time is not as important as are other factors associated with the organic matter. Chief among these is considered to be the rate of "turnover" of organic materials. A high rate of turnover is accomplished when organic materials are added to the soil in large quantity and then decompose quickly. In spite of the lack of correlation between organic matter content and yield, humus is still considered by the author (3) to be necessary for orchard soils.

## PROCEDURE

A total of 74 plots of mature McIntosh trees was selected in grower-owned orchards in the Okanagan Valley in British Columbia, in 1937. In each plot, soil samples were obtained during the months of April and May, 1940. The procedure used was to select a representative tree in each plot, and to make 10 borings with a soil auger around the tree, at distances of 4 to 10 feet from the trunk. The soil was composited at successive depths of 0 to 8 inches, 8 to 24 inches, and 24 to 60 inches. It was then screened through a 3 mm. sieve and allowed to air dry. The procedures used in laying out the plots, taking the tree records over a 6-year period, and obtaining the soil samples, have been outlined in greater detail in previous papers of this series (13, 14).

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The McIntosh plots were selected in 1937 primarily on the basis of tree type, soil texture and soil depth, and little attention was paid to type of cover crop. In addition, no attempt was made to influence the growers in their choice of cover crops or in their methods of handling them.

The only records obtained on the cover crops grown were observational notes, made during the 6 years (1937 to 42) of tree recording. All of the plots were under irrigation, and cover-cropped, and a few were in permanent alfalfa sod, and some in permanent grass sod. These were usually cultivated once a year, in the late fall or early spring. A few plots were in sweet clover, hairy vetch, fall rye, or oats. These were usually cultivated twice a year, once as above and once in midsummer or late summer. In a large proportion of the plots, leguminous cover crops had been planted by the growers, but they had reverted to general mixtures of cover crops, weeds, and grasses. In only two cases was organic matter known to have been applied in other form than as green manure. In both of these cases, some poultry manure had been applied.

Mechanical analyses of the soil samples were made by the hydrometer method of Bouyoucos (2).

The procedure used for determining the organic matter content of the soil was a modification of the hydrogen peroxide method of Robinson (8): weigh a 125 ml. Erlenmeyer flask; add approximately 2 gm. of soil, and weigh again. Add 20 ml. of 3% hydrogen peroxide, and set on steam bath for 30 to 45 minutes. Add another 10 ml. of 3% hydrogen peroxide, washing down the inside of the flask with it. Replace on steam bath for 30 to 45 minutes. Add a further 10 ml. of hydrogen peroxide, place on steam bath and allow to evaporate to dryness. Remove from steam bath, leave near balances for one hour or more to come to moisture equilibrium with the air, and weigh. The loss in weight is considered to represent the amount of organic matter lost by oxidation. The percentage of organic matter thus determined was then adjusted for the percentage of gravel discarded when the soil samples were originally taken, on the assumption that the organic matter content of the gravel was zero.

It was found that the weight of the flasks varied somewhat with the humidity of the air. In order to correct for this, approximately 2 gm. of soil were added to each of the 2 125-ml. Erlenmeyer flasks. These flasks were then weighed every time the other flasks were weighed, so that corrections could be made for variations in air humidity. The error from this source was thereby reduced to below 0.1% of organic matter. No difficulty from manganese was encountered in this investigation.

The above procedure was found to oxidize the organic matter to the point that further additions of hydrogen peroxide did not make any measurable difference in the weight of the soil. This was true with those samples containing the highest amounts of organic matter. It should be pointed out that all of the samples were comparatively low in their contents of organic matter.

As pointed out by Robinson (8), oxidation with dilute hydrogen peroxide does not remove all of the organic matter from the soil. In fact, after oxidation small particles of vegetable matter or coal-like material could, in some cases, still be seen with the naked eye. These residues were



apparently the most resistant portions of the organic matter in the soil. The question arises as to which is the more important measurement, that of *total* organic matter or that of *active* organic matter. In this investigation, it has been assumed that the active portion is the more important. It is realized that segregation of the active portion from the inactive portion is a very difficult matter, and that the procedure used may not be entirely satisfactory in this regard. It appears safe to assume, however, that the results obtained bear a closer relationship to the active organic matter content than to the total organic matter content.

## RESULTS

The terminal lengths and yields of the McIntosh trees in the 74 plots have already been reported for the individual trees (13) and for the plot averages (14). The clay, colloid, and organic matter contents of the soils are summarized in Table 1. Since the records on the soil samples taken below a depth of 24 inches have not been used in the correlations reported in this paper, only the 0 to 8 and 8 to 24 inch data are presented in the table. The 0 to 24 inch percentages of organic matter (last column of Table 1) have been obtained by weighting the figures for the 0 to 8 and 8 to 24 inch samples by their respective depths. Other soils data have been presented in the second paper of this series (14).

Difficulties were encountered in attempting to determine the effects of each kind of cover crop on the organic matter content of the soil and on tree performance. This was due in part to the fact that no measurements were taken of cover crop growth. It was also due to the fact that there were so many different cover crops, and so many combinations of cover crops and fertilizers. Finally, many of the growers had changed their cover-cropping schedules prior to soil sampling. As a result, the relationships between cover crop on the one hand and soil and tree characteristics on the other hand were inconclusive, and will not be reported here.

TABLE 1.—CLAY, COLLOID, AND ORGANIC MATTER CONTENTS OF SOIL SAMPLES

Plot No.	Sand		Silt		Clay		Colloid		Organic matter		
	0 to 8	8 to 24	0 to 8	8 to 24	0 to 8	8 to 24	0 to 8	8 to 24	0 to 8	8 to 24	0 to 24
	%	%	%	%	%	%	%	%	%	%	%
P2	37.4	29.4	46.8	65.4	15.8	15.2	27.2	29.0	2.9	1.3	1.8
P3	39.0	38.2	47.8	51.4	13.2	10.4	24.6	20.6	2.7	1.0	1.6
P4	41.6	27.6	44.0	59.4	14.4	13.0	23.4	30.2	2.6	1.1	1.6
P1	21.2	19.4	62.2	62.8	16.6	17.8	32.2	30.2	1.5	0.6	0.9
P9	52.8	51.0	38.2	35.8	9.0	13.2	17.0	22.2	1.8	1.0	1.3
P10	65.2	64.8	25.0	31.0	9.8	4.2	24.4	8.2	1.2	0.7	0.9
P5	43.4	39.4	42.4	46.0	14.2	14.6	22.2	27.0	2.3	0.9	1.4
P7	55.4	54.4	34.0	31.4	10.6	14.2	19.4	19.4	2.0	0.7	1.1
P6	48.0	54.6	44.8	33.2	7.2	12.2	21.0	18.2	3.2	0.4	1.3
S12	65.2	62.2	25.6	24.4	9.2	13.4	16.0	13.6	1.8	0.2	0.7
S10	67.2	72.2	22.6	20.4	10.2	7.4	15.8	11.0	1.7	0.3	0.8
T2	53.8	41.8	29.8	38.0	16.4	20.2	28.4	35.8	2.2	0.5	1.1
T3	76.8	75.4	16.6	21.6	6.6	3.0	13.6	8.2	1.9	0.1	0.7
T6	47.4	53.4	39.8	36.0	12.8	10.6	24.2	20.6	0.8	0.6	0.7
T7	58.8	61.6	41.0	27.0	10.2	11.4	23.2	16.4	1.6	0.8	1.1

TABLE 1.—CLAY, COLLOID, AND ORGANIC MATTER CONTENTS OF SOIL SAMPLES—*Concluded*

Plot No.	Sand		Silt		Clay		Colloid		Organic matter		
	0 to 8	8 to 24	0 to 8	8 to 24	0 to 8	8 to 24	0 to 8	8 to 24	0 to 8	8 to 24	0 to 24
	%	%	%	%	%	%	%	%	%	%	%
T8	51.6	49.0	29.8	27.0	18.6	24.0	28.2	34.0	1.7	0.7	1.0
T9	73.8	76.0	22.0	19.0	4.2	5.0	11.0	11.0	1.4	0.2	0.6
K1	67.2	62.8	23.4	31.0	9.4	6.2	16.6	11.6	1.4	0.2	0.6
K2	65.4	70.0	24.4	21.8	10.2	8.2	14.6	12.8	1.8	0.6	1.0
K6	64.2	70.2	22.0	22.6	13.8	7.2	18.0	13.2	2.0	0.5	1.0
K21	64.2	70.2	25.0	20.0	10.8	9.8	17.2	15.8	1.7	0.6	1.0
K7	66.6	66.0	21.8	22.8	11.6	11.2	26.8	17.0	1.7	0.8	1.1
K9	56.8	59.6	29.2	30.0	14.0	10.4	21.8	16.4	2.1	0.9	1.3
K27	56.8	68.8	27.0	22.8	16.2	8.4	24.6	12.8	2.1	0.1	0.8
K16	53.8	51.0	33.6	26.6	12.6	22.4	41.6	34.4	2.2	1.4	1.7
K10	55.6	62.8	29.4	26.4	15.0	10.8	20.8	16.6	1.8	0.2	0.7
K39	54.4	58.0	30.8	30.6	14.8	11.4	20.6	28.2	0.8	0.3	0.5
K53	47.8	60.2	33.6	31.0	18.6	8.8	28.4	19.6	2.2	0.5	1.1
K54	54.0	66.4	32.4	23.8	13.6	9.8	20.0	14.4	2.1	0.4	1.0
K11	53.0	70.0	33.4	21.4	13.6	8.6	25.6	12.4	2.7	0.4	1.2
K12	61.2	69.2	24.6	20.2	14.2	10.6	17.6	14.6	2.3	0.2	0.9
K13	57.8	64.2	26.4	29.2	15.8	6.6	24.0	14.2	1.9	0.3	0.8
K14	58.0	67.8	28.2	24.6	13.8	7.6	19.6	12.4	1.9	0.2	0.8
K15	56.2	67.6	32.2	25.2	11.6	7.2	19.8	14.8	1.9	0.4	0.9
K46	53.2	67.2	34.8	22.2	12.0	10.6	22.4	14.8	1.4	0.1	0.5
K51	58.6	65.4	28.6	25.8	12.8	8.8	22.0	15.6	2.0	0.4	0.9
K17	52.2	45.8	24.8	18.0	23.0	36.2	30.8	44.8	3.0	2.9	2.9
K18	25.8	13.0	33.4	24.2	40.8	62.8	50.8	73.0	3.2	2.5	2.7
K22	54.2	64.6	30.8	28.2	15.0	7.2	21.0	14.0	2.1	0.1	0.8
K44	59.2	67.4	27.0	24.8	13.8	7.8	21.4	14.6	2.8	0.5	1.3
K25	19.0	16.2	67.4	21.6	13.6	62.2	50.9	72.2	3.6	3.5	3.5
K24	64.6	73.2	23.0	16.2	12.4	10.6	17.8	15.6	1.3	0.2	0.6
K49	23.8	8.2	28.6	13.4	47.6	78.4	58.4	83.8	3.6	2.5	2.8
K8	43.8	38.2	30.6	26.4	25.6	35.4	34.2	41.4	3.1	1.9	2.3
K48	32.8	13.8	22.0	38.0	45.2	48.2	57.0	63.8	3.8	2.8	3.1
B29	47.2	72.8	44.4	21.0	8.4	6.2	13.2	10.2	1.7	0.7	1.0
B30	61.0	62.6	32.0	28.8	7.0	8.6	35.0	11.2	1.4	0.7	0.9
B31	59.6	59.8	39.4	36.6	11.0	3.6	23.4	6.4	1.8	0.9	1.2
B1	71.6	59.6	18.8	27.8	9.6	12.6	13.0	18.4	2.3	1.2	1.6
B34	65.0	56.0	24.8	33.2	10.2	10.8	16.8	19.2	1.4	1.2	1.3
B33	66.4	75.0	23.6	16.8	10.0	8.2	15.8	10.0	1.1	0.7	0.8
B38	76.8	79.4	16.6	14.6	6.6	6.0	21.6	8.0	1.5	0.8	1.0
B36	68.2	75.8	21.6	19.0	10.2	5.2	15.4	9.0	1.9	0.8	1.2
B37	59.4	66.8	27.8	26.0	12.8	7.2	20.6	12.8	2.0	0.8	1.2
G42	49.0	44.0	44.6	50.0	6.4	6.0	14.0	13.6	1.7	1.3	1.4
G50	64.4	73.2	30.2	22.2	5.4	4.6	11.6	9.2	1.7	0.8	1.1
G26	62.6	56.8	32.4	36.8	5.0	6.4	11.0	11.2	1.2	0.8	0.9
G18	75.8	78.6	17.8	14.0	6.4	7.4	10.8	11.2	1.1	0.3	0.6
G17	59.2	65.8	28.4	19.0	12.4	15.2	20.6	24.2	1.2	0.6	0.8
G19	25.2	20.4	54.4	59.2	20.4	20.4	37.2	42.6	3.6	0.7	1.7
G20	17.6	17.6	42.4	31.6	39.0	50.8	51.6	65.4	3.9	2.5	3.0
W2	54.8	48.6	20.8	14.6	24.4	36.8	29.8	40.0	3.7	3.1	3.3
W7	71.8	76.4	21.8	17.2	6.4	7.4	12.4	12.8	0.6	0.3	0.4
W6	74.0	57.2	16.8	33.0	9.2	9.8	12.8	15.6	1.1	0.3	0.6
W5	73.2	75.8	18.6	16.6	8.2	7.6	14.4	9.8	1.5	0.5	0.8
W4	74.6	73.4	19.6	18.0	5.8	8.6	11.2	11.6	0.8	0.4	0.5
W9	37.4	28.4	33.2	32.6	29.4	39.6	40.4	51.0	3.8	2.6	3.0
W8	65.2	63.4	21.2	21.4	13.6	15.2	19.2	20.4	1.8	0.5	0.9
W10	66.2	71.8	20.2	17.6	13.6	10.6	19.4	14.4	2.2	1.0	1.4
O14	52.2	55.8	29.8	25.2	18.0	19.0	26.2	24.2	2.9	1.1	1.7
O17	68.4	70.0	19.6	18.4	12.0	11.6	18.6	13.8	2.5	1.2	1.7
O15	63.8	72.0	25.0	22.0	11.2	6.0	22.8	12.8	1.2	0.9	1.0
O18	51.8	57.8	28.6	26.0	19.6	16.2	29.8	23.6	2.3	0.6	1.6
O19	54.4	60.2	29.0	25.8	16.6	14.0	27.8	21.8	2.9	0.7	1.4

The general area from which the soil samples were selected ranged from the northern fringe of the brown soils at Penticton to the southern fringe of the black soils at Oyama. It was anticipated, therefore, that the organic matter content would increase from Penticton north to Oyama. An examination of the data has not revealed any such trend. The effects of climate have apparently been masked by the effects of 30 years or more of irrigation, cover cropping, cultivation, and soil erosion.

The usual effects of depth of soil on organic matter content were found. In other words, most of the organic matter was found in the surface 8 inches, and progressively less at the lower depths. A few examples are presented in Table 2.

TABLE 2.—EFFECT OF SOIL DEPTH ON ORGANIC MATTER CONTENT

Plot No.	Soil texture	Organic matter content		
		0 to 8*	8 to 24*	24 to 60*
P2	Silt loam	2.9	1.3	0.9
P10	Sandy loam	1.2	0.7	0.7
K18	Clay	3.2	2.5	0.7
K1	Sandy loam	1.4	0.2	—
G20	Clay	3.9	2.5	1.6
G50	Sandy loam	1.7	0.8	0.5
W9	Clay loam	3.8	2.6	1.3
W4	Loamy sand	0.8	0.4	0.1

\* Depth of soil in inches.

A preliminary examination of the data indicated that the heavier soils contained more organic matter than did the lighter soils. This is illustrated by the four pairs of soils listed in Table 2, one of each pair being a heavy soil and one a light soil. The correlation between the colloid contents and the organic matter contents of the 0 to 8 inch samples of the 74 plots was determined, and found to be +0.653, which was "highly significant" (odds greater than 99 : 1). In other words, there was a tendency for the organic matter to increase as the soil became heavier. In confirmation of this, the correlation between the moisture holding capacity and the organic matter content of the 0 to 8 inch samples was +0.777 and that of the 8 to 24 inch samples was +0.834. Both of these are likewise "highly significant".

The question arises as to whether the high correlations between organic matter content on the one hand and the colloid content and moisture holding capacity on the other hand might be due to the effects of the organic matter content on the other two factors. In other words, the presence of the organic matter will automatically increase both the colloid content and the moisture holding capacity. In order to check up on this, that portion of the moisture holding capacity that could be attributed to the organic matter content was estimated, using 179% as the moisture holding capacity of organic matter. This was based on Olmstead's (7) figure for the normal moisture capacity of organic matter. As an example of the calculation involved, the organic matter content of sample P2 (0 to 8



inches) was 2.9% and that part of its moisture holding capacity due to this organic matter was approximately  $2.9 \times 1.79 = 5.2\%$ . The amounts thus calculated were deducted from the moisture holding capacities (14). The correlation between these residual moisture holding capacities and the organic matter contents was then calculated, and found to be +0.465 for the 0 to 8 inch samples. This is distinctly lower than the original figure of +0.777, but is still "highly significant". It appears from this that there was a strong tendency for the heavier soils to have higher organic matter contents than the sandier soils. The reason for this was not ascertained. It was suspected, however, to be due to at least the following two causes: (a) better growth of cover crops on the heavier soils, resulting in a greater accumulation of organic matter, and (b) greater erosion of the sandier soils, resulting in a greater loss of organic matter.

Included in the 74 McIntosh plots were three series of plots in controlled fertilizer experiments. In these three series, the application of fertilizers containing nitrogen increased the organic matter content above that in the plots receiving no fertilizer, but the addition of phosphate and potash brought about no further increase.

The organic matter content of the soil was correlated with the average terminal length of the trees over a period of 6 years (13). Using the organic matter content in the 0 to 8 inch depth, the correlation was  $-0.007$ ; and using the organic matter in the 0 to 24 inch depth, the correlation was  $+0.103$ . These figures are both "non-significant". In other words, there is no evidence of any relationship between the organic matter content of the soil and the vigour of the trees as represented by terminal length.

Similar correlations were calculated between the organic matter content in the 0 to 8 inch and the 0 to 24 inch depths respectively and the yield of fruit per acre. The yield figures use in making these correlations were those that had been adjusted for differences in size of tree (13). The 0 to 8 inch correlation was  $+0.094$ , and the 0 to 24 inch correlation was  $+0.245$ . This latter figure is "significant" (odds between 19 : 1 and 99 : 1). In other words, there is some evidence of an increase in yield accompanying an increase in organic matter.

The question arises as to whether this increase in yield is due to the presence of the organic matter, or whether it is due to some other factor. In an attempt to determine this, the correlation between the organic matter content of the soil (0 to 24 inch) and the yield of the trees was re-calculated, using a partial correlation to eliminate the effects of differences in soil texture, as represented by the moisture holding capacity. The resulting correlation was  $-0.089$ . The process was then repeated, using the net moisture holding capacity after deducting that portion attributable to organic matter. A correlation of  $+0.123$  was obtained. Neither of these correlations is "significant". There is thus no proof of any relationship between the organic matter content of the soil and the yield of the trees.

## DISCUSSION

Although no proof was obtained from this investigation that organic matter in the soil has a beneficial effect on the growth or yield of tree fruits, it cannot be assumed that there is no such effect. As pointed out by

Cummings (3), the beneficial effects of organic matter may accrue not so much from its mere presence in the soil as from its rate of "turnover". In other words, the decomposition of the organic matter is accompanied by improved bacterial conditions in the soil. The decomposition products, in turn, induce a more rapid breakdown of the insoluble inorganic material. The nature of the organic matter present, its rate of decomposition, and its effects on the chemical and bacterial status of the soil, are not measured by a mere determination of its quantity at any one time.

Another difficulty encountered in this investigation is that other factors than the organic matter content of the soil were active in limiting growth and yield of the trees. By way of example, there was the nitrogen content of the soil. In a high percentage of the plots, both growth and yield were limited by a deficiency of nitrogen. Moreover, this deficiency was in some cases accompanied by a relatively high content of organic matter in the soil, a situation obtained when a permanent grass sod cover crop was grown and an insufficient amount of nitrogenous fertilizer was applied.

Although it has been adequately demonstrated by soil investigators that organic matter benefits the soil in a number of ways, it does not necessarily follow that a high organic matter content is essential to normal crop production. In this present investigation, for example, high yields were obtained from trees in soils whose organic matter contents could not be considered exceptionally high. Plot G26 averaged 1526 loose bushel boxes per acre per year for the 6-year period, yet its soil contained only 1.2% of organic matter in the top 8 inches. The figures for P lot G42 were 1602 boxes and 1.7%; for P lot K24, 1509 boxes and 1.3%; for P lot P9, 1813 boxes and 1.8%; and for P lot P10, 1950 boxes and 1.2%. It is cases like this that have helped to lower the correlation between the organic matter content of the soil and tree yield.

On the other hand, many cases of obvious organic matter deficiency have been encountered. In some of the non-irrigated orchard areas in the southern interior of British Columbia (not covered in this investigation), the rainfall has not been heavy enough to allow the growing of cover crops. Furthermore, stable manure and straw have been expensive, and have been sparingly used. Much of the soil, accordingly, has become too low in its organic matter content. This has resulted in a heavy run-off of the rain or melting snow, especially on silt and clay soils, and all types of soil are now subject to severe surface erosion. Such a condition is seldom encountered in the irrigated areas, where it is a common practice to grow cover crops.

#### SUMMARY

The organic matter content was determined in soil samples from depths of 0 to 8 inches, 8 to 24 inches, and 24 to 60 inches, obtained from 74 plots of mature McIntosh apple trees. Mechanical analyses were also made.

Within the climatic range of the plots, from the northern fringe of the brown soils to the southern fringe of the black soils, no measurable effect of climate on organic matter content was found.

The organic matter content decreased with depth in the soil. It was higher on the average in the silt and clay soils than in the sandy soils. It was increased by applications of nitrogenous fertilizers, but not by applications of fertilizers containing phosphate and potash.

No relationship was found between the organic matter content of the soil at any one time and either tree growth or tree yield.

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# THE OCCURRENCE OF NEW STRAINS OF *PUCCINIA TRITICINA* IN CANADA AND THEIR BEARING ON VARIETAL REACTION<sup>1</sup>

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## INTRODUCTION

With the distribution through much of the spring wheat area in the United States and Canada of Thatcher wheat, leaf rust (*Puccinia triticina* Erikss.) acquired new importance owing to the high susceptibility of this variety to most physiologic races of the rust. Since Thatcher was distributed, a number of hard red spring wheats with more or less resistance to leaf rust have come under cultivation in the same area (e.g., Rival, Pilot, Cadet, Mida, Newthatch, Renown, Regent). The leaf-rust resistance of these and most other recently produced spring wheats was derived from Hope or H-44. As this resistance is manifested to a much greater degree in the adult-plant stage than in earlier growth stages, it is designated as "adult-plant" or "mature-plant" resistance and is accordingly most useful in regions in which leaf rust does not develop until plant growth is well advanced.

For several years after the distribution of wheat varieties with adult-plant resistance it appeared that this type of leaf-rust resistance was highly satisfactory, at least under conditions prevailing in Canada. Varieties such as Renown and Regent rarely showed more than 10 or 15% infection, and losses caused to these varieties were inappreciable. Tests made in the greenhouse in the spring of 1941 indicated (7) that Renown and Regent possessed adult-plant resistance to all of the 19 physiologic races employed, namely, races 1, 2, 3, 15, 20, 27, 28, 29, 31, 34, 39, 44, 52, 58, 71, 83, 89, 104, and 130. As a result of these tests it was concluded that "it seems probable that these two wheats exhibit towards North American races of leaf rust a resistance as general though not as great as the resistance they show to physiologic races of *Puccinia graminis Tritici* Erikss. and Henn."

## RECENT PATHOGENIC CHANGES IN LEAF RUST

Although Renown and Regent displayed, in general, a high resistance to leaf rust there occurred now and then isolated instances of an apparent breakdown in resistance. For example, in 1936, Regent (R.L. 975.1) growing in the rust nursery at Ottawa, Ont., bore a 50% infection; and in 1937, Renown (R.L. 716) at Agassiz, B.C., bore a 75% infection while at other stations infections of 30 to 50% were not uncommon. As no unusual physiologic races were detected in connection with these outbreaks, the severity of infection was attributed to the influence of environmental conditions.

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Despite such sporadic evidence of breakdown in the leaf-rust resistance of Renown and Regent, little concern was felt for their rust reaction until the summer of 1943 when Regent, Renown, and other derivatives of Hope and H-44 rusted severely in the field plots at Winnipeg under conditions of an artificially induced epidemic in which all available physiologic races were present. In the same summer, severe infection of Regent and Renown (60 to 80%) was recorded in uniform rust nurseries at Ottawa and Manotick, in eastern Ontario.

As the physiologic races isolated from these 3 localities were also generally present in other localities where no breakdown in resistance occurred, it was again concluded that unusual environmental influences had affected varietal reaction—an inference that seemed reasonable in view of the fact that the severe infection was present chiefly on grain sown unusually late.

In the summer of 1945, there was for the first time evidence of a widespread breakdown in the leaf-rust resistance of Regent and other varieties with similar reaction (*vide* Tables 1 and 2). Throughout Manitoba and eastern Saskatchewan, and in many localities in Eastern Canada, Regent reacted like a moderately susceptible variety. At the same time there was a sharp increase in the number of isolates of physiologic race 128, a race that, at least under the greenhouse conditions prevailing at Winnipeg, is scarcely distinguishable from race 29. Race 128 was first identified, in Canada, in 1944. In that year, it made up 11.8% of all leaf rust isolates and in 1945 this rose to 26.1%. However, this race was, undoubtedly, less prevalent than is indicated by the above percentages because many collections were made on varieties that tended to select out this race. The most notable feature of the physiologic race survey of 1945 was the fact that race 128 accounted for no less than 66% of the isolates derived from Regent, Renown, and Coronation (a variety with adult-plant resistance widely grown in Eastern Canada).

TABLE 1.—AVERAGE PERCENTAGES OF LEAF RUST ON REGENT (R.L. 975.6) AND THATCHER (R.L. 1246) IN UNIFORM RUST NURSERIES IN CANADA, 1940 TO 1945.  
(ONLY THOSE STATIONS ARE INCLUDED IN WHICH MORE THAN  
A TRACE OF LEAF RUST WAS PRESENT ON REGENT)

Year	1940	1941	1942	1943	1944	1945
No. stations	7	12	12	12	15	21
	%	%	%	%	%	%
Regent	9	9	12	24	17	52
Thatcher	64	60	86	82	67	85

TABLE 2.—AVERAGE PERCENTAGES OF LEAF RUST ON WHEAT VARIETIES IN CO-OPERATIVE TESTS AT WINNIPEG, MORDEN, AND BRANDON, MANITOBA, 1942 TO 1945

Year	1942	1943	1944	1945
	%	%	%	%
Regent (R.L. 975.26)	10	19	15	52
Regent × Thatcher (R.L. 2038)	9	19	13	54
Cadet (R.L. 1597)	7	14	17	51
Newthatch (R.L. 2752)	—	19	26	52
Regent (R.L. 975.6)*	10	28	40	80
Thatcher (R.L. 1246)	67	75	64	79

\* Percentages from uniform rust nurseries at same stations but sown later.

At this point, a discussion of the relationship between races 29 and 128 is advisable. Race 29 had been identified from field collections annually since 1937 but was regarded as of minor significance for 2 reasons. First, it occurred relatively infrequently; and second, it was no more virulent on Renown and Regent than other races (7). In 1944, race 128 was identified for the first time but the reactions of the differential hosts to this race and to race 29 were so similar that there was frequently some hesitation as to whether a particular culture should be identified as one race or the other. In that year, race 29 made up 7.7% and race 128, 11.8% of all isolates—a combined total of 19.5%. In the leaf rust survey of 1945, similar difficulties were experienced in distinguishing the 2 races. Because of these difficulties, all cultures suspected of being one or the other of these 2 races were provisionally designated as race 29/128. Owing, however, to the frequent occurrence of this pathogenic type of rust in collections made on Renown and Regent, many of the isolates so designated were kept for infection tests with adult plants of Regent wheat.

In these infection tests, performed in the greenhouse from November, 1945 to March, 1946, it was found that 14 of 18 cultures of the race group designated as 29/128 infected Regent so severely that this variety had to be regarded as completely susceptible, whereas 4 cultures infected it rather lightly (Table 3). It seemed reasonable, therefore, to suppose that the 4 cultures that attacked adult Regent plants lightly corresponded to the race 29 collected in former years, whereas the 14 cultures that attacked Regent plants severely represented a pathogenic strain uncommon in former years. This latter strain was regarded as race 128 because that race was first identified at a time when Regent wheat was apparently losing much of its former resistance; but, owing to the difficulty of distinguishing it from race 29, it might with equal propriety be regarded as a biotype of that race. It was also decided to identify as race 128 those cultures of the race 29/128 group that were not included in the greenhouse tests with mature plants but were collected on heavily infected Regent or varieties with similar reaction.

TABLE 3.—PATHOGENICITY OF CULTURES OF 14 PHYSIOLOGIC RACES OF LEAF RUST TOWARDS ADULT PLANTS OF REGENT WHEAT

Physiologic race	No. cultures tested	No. cultures attacking Regent heavily	No. cultures attacking Regent lightly
2	1	0	1
3	3	0	3
5	5	3	2
9	6	0	6
11	1	0	1
15	11	2	9
29/128	18	14	4
34	1	0	1
58	2	0	2
65	2	1	1
76	4	0	4
101	1	1	0
104	2	0	2
113	2	0	2



Although race 128 was first identified, in Canada, in 1944, there is evidence of its presence at an earlier date. In the above-mentioned tests with adult Regent plants, there was included a culture collected at Morden, Manitoba, in 1943 and at that time identified as race 29. This culture proved highly pathogenic to Regent and therefore must now be assigned to race 128 rather than race 29.

Although race 128 is by far the most widely prevalent strain of leaf rust capable of attacking Regent, it is by no means the only one. Of the races used in the tests with adult plants (Table 3), Regent was attacked heavily by 3 out of 5 cultures of race 5, by 2 of 11 cultures of race 15, and by 1 out of 2 cultures of race 65. It was also attacked heavily by the 1 collection of race 101 used in the tests. It may be mentioned here that the 2 last-named races bear a considerable resemblance to races 29 and 128.

It should be clear from the above discussion that the differential hosts now used for differentiating leaf-rust races are not adequate to indicate to the investigator the pathogenic changes taking place in the rust. The changes that have taken place were not apparent because may of the new strains pathogenic to Regent and related wheats were merely biotypes of races that had been present in past years. It seemed necessary to conduct a search for supplementary hosts that would indicate by their seedling reactions the pathogenicity of the rust towards wheat varieties that are now widely grown. The investigator should be able to tell by means of seedling reactions whether any given culture of rust will attack the adult plants of a given wheat variety of economic importance. The results of tests in which a comparison was made of seedling and adult-plant reaction show that rust cultures that attack Regent, Redman, and Renown heavily in the adult stage cannot be distinguished, by the seedling reaction of Regent wheat, from those that attack these varieties lightly in the adult stage. The reaction of Regent will not serve this purpose because of the rather uniform susceptibility of the seedlings to all cultures used in the test. In both Renown and Hope, however, there was a relation between seedling and adult-plant reaction, both varieties being susceptible in the seedling stage to those cultures that attacked adult plants of Regent and Renown heavily, and moderately resistant (showing X type of infection) to all cultures that attacked Regent and Renown lightly in the adult-plant stage. Hence it is probable that either Renown or Hope may serve as a supplementary differential host for distinguishing cultures that attack adult plants of Regent and Renown severely. The utilization of such a supplementary differential host to distinguish 2 strains that are undistinguishable by the standard differential hosts is not a new development. In Australia, the variety Thew has been used by Waterhouse (8) to distinguish certain strains of leaf rust that produce identical infections on the standard hosts.

#### RESISTANCE TO NEW PATHOGENIC TYPES OF LEAF RUST

Field observations and greenhouse tests alike indicate that the apparent loss of leaf-rust resistance in Regent wheat also applies to other varieties derived from Hope and H-44. In the leaf rust nursery at Winnipeg, in 1943 and in 1945, the varieties Renown, Cadet, and Newthatch bore approximately the same percentage of rust as Regent and, in general, gave

the same impression of susceptibility. The variety Hope, though not included in the leaf rust nursery, was grown in nearby plots and appeared to be susceptible though perhaps less so than Regent. It would seem, therefore, that the adult-plant resistance of Hope, H-44, and their derivatives is no longer fully effective, at least in seasons favourable to leaf rust epiphytotics.

Fortunately there are available leaf-rust-resistant wheat varieties that do not appear to have lost any of their resistance with the advent of new strains of the rust and, although these varieties may not in themselves be of commercial value, they are nonetheless valuable for breeding purposes.

Among varieties that in 1943 and 1945 were grown in the field plots at Winnipeg, and there maintained the high leaf-rust resistance they had shown in previous years, may be mentioned Chinese  $\times$  Marquis (R.L. 1596), K-33 (R.L. 1885), and Warden  $\times$  Hybrid English W325. The resistance of Chinese  $\times$  Marquis and K-33 is confined to the adult plant, the seedling reaction being at least moderately susceptible. The resistance of Warden  $\times$  Hybrid English W325, on the other hand, is uniformly the same in all stages of plant growth (6, 7).

TABLE 4.—REACTIONS, IN THE GREENHOUSE, OF CERTAIN WHEAT VARIETIES IN THE ADULT STAGE TO PHYSIOLOGIC RACES OF LEAF RUST

Physio- logic race	Place of collection	Year	Renown R.L. 716.6	Regent R.L. 975.6	Redman R.L. 1834.1	Hybrid R.L. 2325	Hybrid R.L. 2327	Frontana	Fronteira
3	Lang, Sask.	1945	—	MR	MR	R	R	—	—
3	Fredericton, N.B.	1945	MR	MR	MR	R	R	VR	VR
5	Melita, Man.	1945	S	S	S	R*	R	VR	R
5	Indian Head, Sask.	1943	—	MR	R	R	R	—	—
9	Lacombe, Alta.	1945	—	MR	MR	R	R	—	—
9	Lethbridge, Alta.	1945	MR	MR	MR	R*	R	VR	R
9	Guelph, Ont.	1945	—	MR	MR	R	R	—	—
9	Kapuskasing, Ont.	1944	—	MR	MR	R	R	—	—
9	Morden, Man.	1942	—	MR	MR	R	R	—	—
11	Fredericton, N.B.	1945	MR	MR	MR	R	R	—	—
15	Gordon Head, B.C.	1945	MR	MR	MR	MR	MR	VR	VR
15	L'Assomption, Que.	1945	—	MR	MR	R	R	—	—
15	Duff, Sask.	1945	MS	S	S	R*	R	VR	VR
15	Indian Head, Sask.	1945	—	MR	MR	R	R	—	—
34	Duhamel, Alta.	1943	—	MR	MR	R	R	—	—
58	Ste. Anne de la Pocatiere, Que.	1945	—	MR	MR	MR	MR	—	—
58	Fredericton, N.B.	1945	—	MR	MR	MR	R	—	—
65	Scott, Sask.	1944	—	MR	MR	R*	R	—	—
65	Saanichton, B.C.	1945	—	S	S	R	R	VR	R
76	Macdonald College, Que.	1944	—	MR	MR	R	R	—	—
101	Altamont, Man.	1945	S	S	S	R	R	VR	VR
104	Bagot, Man.	1943	—	MR	MR	R	R	—	—
113	Guelph, Ont.	1943	—	MR	MR	R	R	—	—
128	Stockholm, Sask.	1945	—	S	S	R	R	VR	MR
128	Deloraine, Man.	1945	S	S	S	R*	R	VR	R
128	Margo, Sask.	1945	—	S	S	R	R	—	—
128	Souris, Man.	1945	S	S	S	MR	R	—	—
128	Saskatoon, Sask.	1945	S	S	S	R	R	—	—
128	L'Assomption, Que.	1945	—	S	S	R	R	VR	MR
128	Morden, Man.	1943	—	S	S	R*	R	—	—

\* Variety not pure—Some plants MR.

Explanation of symbols: VR = very resistant; R = resistant; MR = moderately resistant; MS = moderately susceptible; S = susceptible.

It is evident from recent greenhouse tests that the type of resistance possessed by Warden  $\times$  Hybrid English W325 is not affected by the presence of strains of leaf rust that have overcome the adult-plant resistance of the Hope-H-44 type. Two hybrid lines, R.L. 2325 and R.L. 2327, derived from the cross [McMurchy  $\times$  (Warden  $\times$  Hybrid English W325)]  $\times$  Redman, made at the Dominion Laboratory of Cereal Breeding, Winnipeg, were tested in the adult stage, in March, 1946, for their reactions to a number of leaf-rust cultures capable of attacking both Regent and Redman heavily. The reactions of the hybrid lines as well as the reactions of Regent and Redman, obtained at the same time, are shown in Table 4. Included in this table are also the adult-plant reactions of Renown and 2 wheats, Frontana and Fronteira, seed of which was recently obtained from the Argentine through the kindness of Mr. A. R. da Silva.

The data presented in Table 4 show clearly that the resistance derived from Warden  $\times$  Hybrid English W325 is not affected by those races that attack Regent and Redman heavily. In R.L. 2327, where this type of resistance was most clearly manifested, the reaction of the several leaves of the same plant was nearly uniform. Generally, the flag leaf showed small, necrotic flecks and a few, minute, type-1 pustules (Figure 1). On the second and third leaves from the top, flecks were fewer and type-1 pustules more frequent; and on the fourth and fifth leaves, flecks were few and type-1 pustules common and sometimes numerous. Despite the high resistance indicated by the type of pustule present, it is possible that the sharp necrosis

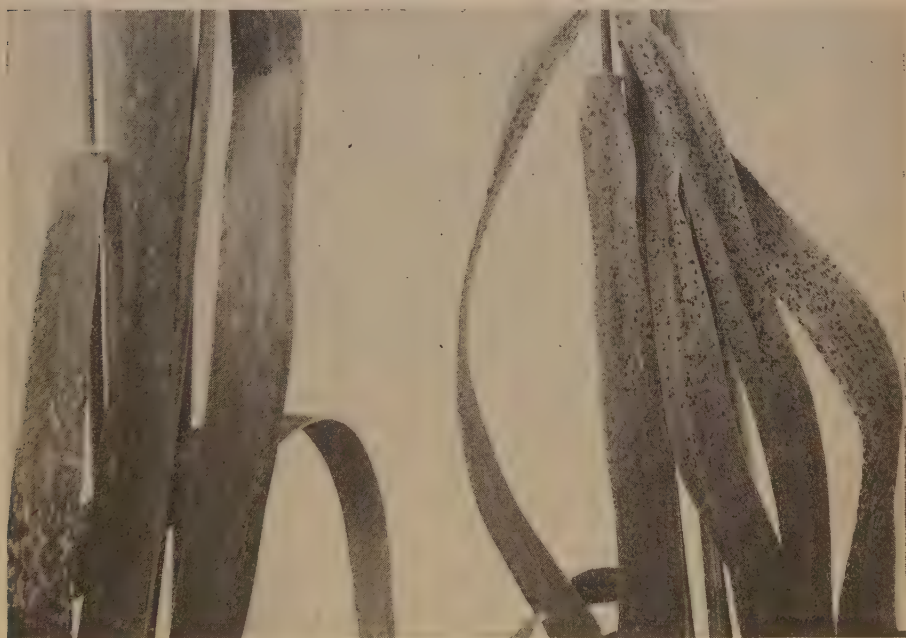


FIGURE 1. Leaf rust infection produced by race 128 on uppermost leaves of adult plants of 2 wheat varieties.

Left—Hybrid line (R.L. 2327) of the cross [McMurchy  $\times$  (Warden  $\times$  Hybrid English W325)]  $\times$  Redman.

Right—Regent (R.L. 975.6).



surrounding the pustules might cause appreciable damage to heavily infected plants. Under exactly the same greenhouse conditions, Regent and Redman showed towards races to which they were susceptible in the adult stage a uniform susceptibility on the different leaves of the same plant. Towards races to which they were resistant in the adult stage they manifested a gradation of reaction: high resistance (flecks and type-1 pustules) on the flag leaf, moderate resistance (2 or X type) on the second and third leaves, and moderate susceptibility (type-3 pustule) on the fourth and lower leaves.

Frontana and Fronteira showed in the adult stage an even higher resistance than did R.L. 2327 (Table 4). In Frontana, this resistance was so great that it approached immunity—numerous small necrotic flecks but few pustules being present. The resistance of these varieties to the cultures that attacked Regent heavily appeared to be much the same as to the other cultures. Reactions of Frontana seedlings to the same cultures showed that seedling resistance in this variety is definitely of a lower order than that of the adult plant. Whereas adult-plant reaction in this variety was in all cases classed as very resistant, seedling reaction varied from resistance (to races 3 and 15) to moderate resistance (to races 9 and 128). Insufficient seed of Fronteira was available to allow this variety to be tested in the seedling stage to individual races; but a single test with a mixture of several races indicated a moderately susceptible seedling reaction.

Tests that have been carried out thus far at Winnipeg, on the seedling reaction of wheat varieties, do not indicate that there are many varieties of *Triticum vulgare* that have general resistance to the physiologic races now prevalent in Canada. Out of 30 *vulgare* varieties tested in the spring of 1945 for their reaction to races 1, 3, 5, 6, 9, 15, 65, 76, 104, 113, 126, and 128, only one variety, Warden  $\times$  Hybrid English W325, possessed high resistance to all the races. In the spring of 1946, more than 40 other varieties belonging to several species of *Triticum* were tested to 29 leaf-rust cultures, mostly 1945 collections, comprising races 5, 9, 15, 58, 76, and 128. Of the 22 *vulgare* wheats used only one, the Argentine wheat La Prevision 25, displayed high resistance to all of the cultures. No tests have been carried out with adult plants of this variety but presumably it is resistant also in the adult stage. Of the non-*vulgare* wheats included in the tests, the following showed high resistance to all of the leaf-rust cultures: *T. monococcum* var. *flavescens* (2 strains, one of which was the named variety Einkorn); *T. turgidum* var. *mirabile*; *T. polonicum* var. *Halleri*; *T. durum* var. *libycum*; and *T. aegilopoides*.

#### EFFECT OF TEMPERATURE ON VARIETAL REACTION TO LEAF RUST

Not the least among the considerations to be taken into account in evaluating the importance of any particular type of leaf-rust resistance is its stability with regard to the environment in which the plant is growing. Conditions of temperature, moisture, and light undoubtedly influence, to a certain extent, varietal reaction to leaf rust. As temperature is thermostatically controlled in certain of the greenhouses of the Dominion Laboratory of Plant Pathology, at Winnipeg, it was possible to test the influence of this factor on leaf-rust development on several wheat varieties with different types of rust reaction.

Of the 7 varieties chosen for the tests, Marquis, McMurachy, and Thatcher are moderately to highly susceptible in all growth stages; K-33, Chinese, and Hope are more or less susceptible in the seedling stage but possess resistance in the adult stage; whereas Warden  $\times$  Hybrid English W325 is resistant in all growth stages.

Four physiologic races—5, 9, 15, and 76—were used and 2 tests were performed, one in January and one in April, 1943. In both tests, plants were grown to heading under ordinary greenhouse conditions. After inoculation, the plants were placed in two thermostatically-controlled greenhouses maintained at approximately 60° F. and 80° F., respectively—half the plants inoculated with each race of the rust being placed in each greenhouse.

The results of the tests (Table 5) show that varieties susceptible at ordinary temperatures may become resistant at high temperatures. The critical temperature for the induction of resistance in a susceptible variety appears to be slightly above 80° F.; earlier experiments (3) had indicated a temperature of 85° F. or higher. It is likely that the exact temperature level at which this change takes place is influenced by other factors in the environment. It is perhaps worthy of note that the temperature level at which McMurachy wheat became resistant to leaf rust is almost exactly that at which it has been shown to become susceptible to stem rust (5).

The reactions of Hope and Chinese wheats were less stable than that of the other variety with adult-plant resistance, *i.e.* K-33. Conditions other than temperature may have exerted an influence as both Hope and Chinese were more resistant in the April test than in the one performed in January. Possibly the conditions of daylight in the April test were more conducive to the development of resistance than the poorer light conditions of the January test. If so, the influence of light on the leaf-rust reaction of Hope and Chinese is much the same as on the stem-rust reaction of Hope (2, 4) which is most susceptible under conditions of short day and diffuse light.

The reaction of Warden  $\times$  Hybrid English W325 was influenced very little by temperature or by the different conditions of light prevailing in the two tests.

### DISCUSSION

In view of the extensive cultivation in Canada and the United States of wheat varieties with the Hope-H-44 type of adult-plant resistance to leaf rust, the breakdown of this resistance would be a matter of considerable economic importance. There is not yet sufficient evidence to state categorically that this type of resistance will no longer be effective in future years. It must be admitted, however, that varieties with this type of resistance gave the impression of moderate susceptibility in 1945 in Manitoba and eastern Saskatchewan.

The future reaction to leaf rust of such varieties is a matter of conjecture because it is not known whether the new pathogenic strains will persist in coming years nor is it known whether the loss of resistance in these varieties in 1945 was due entirely to the presence of these strains of the rust or was in part due to environmental effects. It may be that these varieties will rust severely only in years in which conditions are particularly favourable to leaf rust development, but it should be noted



TABLE 5.—REACTION, IN GREENHOUSE TESTS, OF 7 WHEAT VARIETIES TO 4 PHYSIOLOGIC RACES OF LEAF RUST AT DIFFERENT TEMPERATURES

Variety	Time of test	Temp. °F. (Mean)	Race 5		Race 9		Race 15		Race 76	
			Adult	Seedling	Adult	Seedling	Adult	Seedling	Adult	Seedling
K-33 (R.L. 1885)	Jan.	77	R	MS	R	MR	I to R	MS	R	R
	Apr.	59 82 61	I to R R R	MS — —	I to R I R	MS — —	I to R R R	MS — —	R R R	MS — —
Chinese (R.L. 1815)	Jan.	79	MR to MS	MR	MR	MS	MR	MS	MR to MS	MS
	Apr.	58 82 61	MR R MR	MS — —	I to R I to R	MS — —	MR R MR	MS — —	MR R MR	MS — —
Marquis (R.L. 84)	Jan.	78	MS	MS	R to MR	MS	MS	MS	MS	MR
	Apr.	58 82 61	MS R MR	MS — —	MR I to R R	MS — —	MS R R	MS — —	MS R MR	MS — —
Warden × Hybrid English (R.L. 1803)	Jan.	78	I to R	R	I to R	I	I	I	I to R	R
	Apr.	58 82 61	I to R I to R I	I — —	I to R I I	I — —	I I I	I — —	I to R I to R I	R — —
McMurachy (R.L. 1313)	Jan.	79	MS	MS	MS	MS	MS	MS	MS	MS
	Apr.	57 82 61	MS R MS	MS — —	MS R MS	MS — —	MS R MS	MS — —	MS R MS	MS — —
Thatcher (R.L. 1945)	Jan.	79	MS	MS	MR	MS	MS	MS	MS	MS
	Apr.	58 82 61	MS R MS	S — —	MR to MS I to R R	MS — —	MS R MS	MS — —	MS R MS	MS — —
Hope (R.L. 209)	Jan.	83	MR to MS	MS	MR	MS	MR to MS	MS	MR to MS	MS
	Apr.	58 82 61	MR R I to R	MR — —	MR I to R I to R	MR — —	MR R MS	MS — —	MR R R	MS — —

Explanation of symbols: I = immune; R = resistant; MR = moderately resistant; MS = moderately susceptible; S = susceptible.



that Regent and Renown wheats now show complete susceptibility in the greenhouse to some strains of the rust while they still retain their usual moderate or even high resistance to other strains under identically the same environmental conditions. Although greenhouse infection tests can never be accepted as an exact index of field reaction, this fact suggests strongly that the susceptibility of these varieties in the field is due to the presence of pathogenic strains adapted to them rather than to environmental conditions.

As far as it is possible to judge from greenhouse and field tests at Winnipeg, it would seem that the leaf-rust resistance of Warden  $\times$  Hybrid English W325 is unaffected by the new rust strains highly pathogenic to Regent. If so, this variety will have definite value to plant breeders concerned with the development of leaf-rust-resistant wheats. In view of the great variability in the pathogenicity of leaf rust it is probably too much to expect this variety, or perhaps any other, to remain resistant indefinitely. Recent information from South America (1) shows Warden  $\times$  Hybrid English W325 to be susceptible to physiologic race 5, presumably a collection of it made in the Argentine. This is further evidence of the presence of biotypes in races of the leaf-rust organism, as the Warden  $\times$  Hybrid English W325 type of resistance has proved highly effective against Canadian collections of race 5 (Tables 4 and 5).

The infection tests with adult plants of the varieties Frontana and Fronteira and the seedling tests with the South American wheat La Prevision 25, indicate further possibilities for the breeding of leaf-rust-resistant wheats suitable to Canadian conditions. Though no infection tests have been made with adult plants of La Prevision, it is probable that its resistance in the adult stage is at least equal to its resistance in the seedling stage. That this variety may possess resistance to many physiologic races is suggested by a report from the Argentine (1) that it proved resistant to the 6 races to which it was tested, namely, races 5, 20, 49, 57, 62, and 114. This variety may prove of some value as breeding material for the production of leaf-rust-resistant spring wheats.

In conclusion, it must be emphasized that in view of the pathogenic changes that have obviously been taking place in leaf rust in recent years it is imperative that adequate annual surveys be conducted to determine the physiologic races present each year. Not only must these races be identified but it is equally essential that a study be made of their pathogenicity towards varieties now in cultivation and, further, that a search be made for resistant wheat varieties that may serve as breeding material in case the need for such arises.

#### SUMMARY

It has been shown that there are now present in Canada strains of leaf rust that are capable of heavily attacking Regent wheat and other varieties with similar reaction to this rust. Some of these strains must be regarded as biotypes of known races such as races 5 and 15 but the one most commonly present is identified as race 128. This race bears a close resemblance to race 29, which has been found in Canada annually since 1937, but differs by its ability to rust Regent, Renown, and Redman severely in the adult-plant stage. Race 128 was first identified, in Canada, in 1944 when it



comprised 12% of all leaf rust isolates, but there is evidence of its presence in the preceding year. In 1945, this race comprised 26% of all leaf rust isolates and 66% of those derived from Regent, Renown, and Coronation.

The presence of the above-mentioned strains of leaf rust appears to be the chief reason for the severe outbreak of this rust that occurred in 1945 on Regent and other wheats with similar rust resistance. It probably also accounts for sporadic outbreaks of leaf rust on these varieties in the 2 preceding years.

The new strains of leaf rust that seem to have overcome the resistance of Regent wheat have not affected the resistance of certain other wheats particularly K-33, Chinese  $\times$  Marquis, and Warden  $\times$  Hybrid English W325. Hybrid lines derived from the cross [McMurachy  $\times$  (Warden  $\times$  Hybrid English W325)]  $\times$  Redman show rather high resistance to all leaf-rust races to which they were tested, including strains of the rust capable of attacking Regent and Redman severely. The South American varieties Frontana, Fronteira, and La Prevision 25 also proved highly resistant to all leaf-rust strains to which they were tested.

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